Vol. 11 August, 1957 No. 1

JOURNAL OF THE FACULTY OF AGRICULTURE KYUSHU UNIVERSITY

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PUBLISHED BY THE FACULTY
FUKUOKA
1957

Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11, No. 1
August 30, 1957

On the occurrence of aneuploidy in the offspring of the artificially induced auto-tetraploid plants in Japanese radish (*Raphanus sativus* L.) and Chinese cabbage (*Brassica pekinensis* Rupr.)

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Introduction

Since the establishment of the colchicine technique a large number of the auto-polyploid plants or strains have been raised artificially in various cultivated species, and studies on the character and behaviour of those polyploid plants or strains have hitherto been to some extent accomplished. Excepting only few cases, with most examples we have not been, however, quite successful in obtaining results of certain practical value. Such a situation is considered to have been effected through the following circumstances which prevail on the breeding procedures accompanying chromosome doubling: that there usually exist urgent needs for preparing a large number of test strains covering many relating garden varieties, and also for rather many years of painstaking work on practical breeding (See Fukushima, Tokumasu and Oguro, 1949); that the auto-polyploid plants raised already in various crops have shown certain difficulties in the maintenance of their exact polyploid nature throughout successive generations. Moreover, with some of those tetraploid plants somewhat easy reduction of chromosomes towards the original diploid state during the course of sexual reproduction has also been reported (e.g., Kondo and Karitani (1947) in rice; Noguchi (1950) in *Helianthus annuus* L.). The present authors have also noticed not infrequently the spontaneous occurrence of a similar phenomenon in some strains of auto-tetraploid raised in several varieties

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of Cruciferous vegetables. The exact nature of those situations is yet quite obscure. To get a clue towards solving the problems concerning it may be a primary necessity in order to obtain clear insight into the chromosomal behaviours as well as the breeding procedures of those polyploid individuals or strains.

The present report covers the results of cytogenetic studies carried out with the polyploid individuals, including some hyper- and hypoploid ones, obtained in the offspring of the auto-tetraploid individuals which have been colchicine-induced by the senior author some years ago in the strains of Japanese radish and Chinese cabbage.[1]

Material and method

The auto-tetraploid strains of Japanese radish 'Osaka-Shijûnichi', 2n=36, and that of Chinese cabbage 'Tsujita', 2n=40, both of which were raised artificially by the senior author, were used as the materials.[2] These polyploid strains were maintained throughout several generations after their origin under the controlled pollinations or the selfing through bud-pollination technique. As compared with the normal diploid seeds, the seeds from autotetraploid individuals were clearly larger in size and took more or less uniform appearance, excepting a certain amount of abnormal seeds which occurred along with the normal large ones, but took much smaller sizes than the diploid seeds. The frequency occurrence of those abnormally small seeds varied among the individuals belonging to the same strain, and also showed a certain variation year after year. As well be made clear elsewhere, it was ascertained that those abnormally small seeds from the tetraploid individuals are autotriploid in nature.

The tetraploid individuals used in the present investigations were chosen from the strains which showed rather high occurrence of the exceptional seeds; i.e., Plant No. 3 (1950–51) in Japanese radish, *Raphanus sativus* L., and Plant No. 6 (1949–50) in Chinese cabbage, *Brassica peckinensis* Rupr., were used as the mother plants, the offspring of which were treated exclusively in the present report. Seeds produced

[1] The present work was undertaken as a part of the general inquiries on the problems of seed growing in vegetable crops. The authors gratefully acknowledge the subsidies given from the Scientific Research Fund of the Ministry of Education.

[2] Since the season of 1937–38 a large number of the autotetraploid strains in Japanese radish and several *Brassica* species have been colchicine-induced by Fukushima, and some of those autopolyploid strains have hitherto been maintained and used as the materials for various studies. The two strains treated in the present investigation also have their origin among the first auto-tetraploid forms grown in the season of 1939–40.

under the open-pollination were collected from each individual, and their sizes were compared. Radish seeds were sown at 17th Sept. 1951 and the cabbage seeds at 13th Nov. 1951. The diploid seeds were sown as the controls at the same date as the polyploid ones. Examinations upon certain morphological characters were carried out with the individuals grown up from those seeds. With a part of those individuals the meiotic behaviours of chromosomes in their pollen mother-cells were observed, and their somatic chromosome numbers were ascertained at the same time. Cytological studies were carried through exclusively on the acetic-orcein smear preparations of the pollen mother-cells.

Results of observation

I. Size of seed and other morphological characters

The longer diameters of seeds obtained from the radish Plant No. 3 varied markedly as shown in Fig. 1, which was compiled on the data

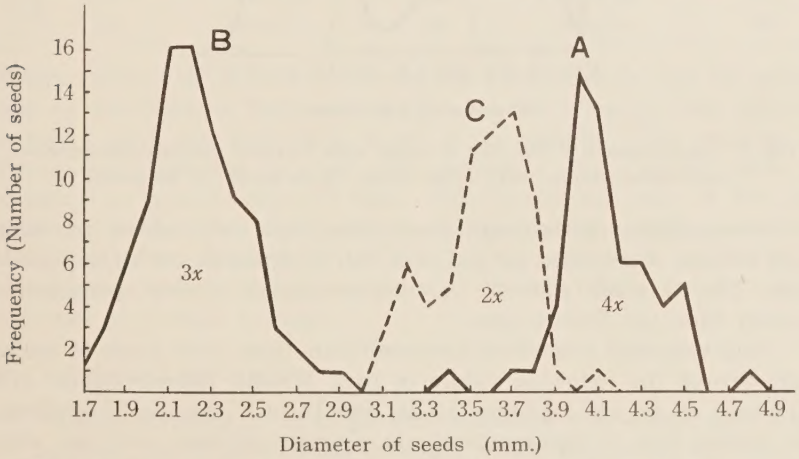


Fig. 1. Comparisons of the sizes of seeds with Japanese radish plants in a polyploidal series. N.B. A, 4*x* seeds; B, 3*x* seeds; C, 2*x* seeds.

of measurements with 231 seeds in total. They were dispersed into two distinct polygons; i.e. A, consisted of 144 seeds, and B, consisted of 87, whose mean values were 4.21 and 2.22 mm. respectively. Fig. 2 represents similar situation of seeds produced on the Plant No. 6 in Chinese cabbage. Two hundred and thirty-two seeds were distributed into two distinct polygons, each consisted of 141 (A) and 91 (B) respectively, their mean diameters being 2.20 and 1.36 mm. In both the species, the seeds showing larger mean diameters are considered to correspond to the tetraploid ones, and the rest with smaller diameters to the triploid.

The polygons denoted with C in both figures were compiled from the data of diploid seeds respectively.

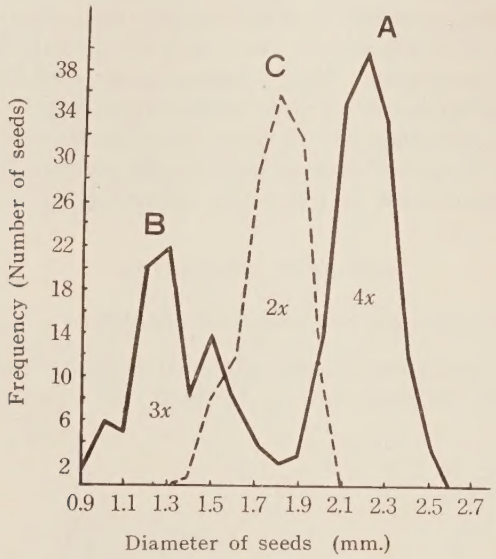


Fig. 2. Comparisons of the size of seeds with Chinese cabbage plants in a polyploidal series. N.B. A, 4*x* seeds; B, 3*x* seeds; C, 2*x* seeds.

Germinability of the small seeds, which are well defined as being 3*x* in nature, was rather normal and well comparable with the diploid ones. The 4*x* seeds showed, in turn, somewhat weaker germination capacity than the diploid ones.

Fully expanded cotyledons developed from those three kinds of radish seeds showed the following values in their breadth measurements: 27.0 $\pm$1.4 mm. in 4*x*, 11.7$\pm$1.8 mm. in 3*x*, and 19.0$\pm$1.3 mm. in 2*x* seedlings. The similar data of measurements on Chinese cabbage seedlings were as follows: 27.0$\pm$2.2 mm. in 4*x*, 13.1$\pm$1.8 mm. in 3*x*, and 21.5$\pm$3.2 mm. in 2*x* seedlings respectively.

The first foliar leaves on the 4*x* seedlings showed most rapid expansion as compared with those of 2*x* or 3*x*. The 3*x* seedlings expanded their first leaves rather slowly, but they realized the most vigorous growing afterwards. Thus the number of leaves expanded before a certain date had shown clear correspondence to the mode of growing in each seedling, representing its developmental velocity. For examples, until 22nd Jan. 1952 4*x* seedlings had developed 28.3 leaves in average, 3*x* ones 30.4 leaves, and the normal 2*x* ones as many as 40.9 leaves. But the blooming had begun earliest in 2*x* individuals, being followed by 4*x* ones, and that in 3*x* ones had fallen behind the

former two, sharply concurring to the growth pattern and vigour of the plants with different nuclear contents.

To compare the size of cells, the major diameters of stomatal guard-cells on the undersurface of fully expanded leaves were measured. The number of chloroplasts in each guard-cell was also counted as another indicator of nuclear structures (See Table 1). Cells of an in-

Table 1. Comparison of major diameters of stomatal guard-cells and number of chloroplasts in each cell.

Polyploidy		Average diameter of guard-cells (micrometer unit)	Number of cells measured	Average number of chloroplasts in each cell	Number of cells observed
Japanese radish	$4x$	19.2±2.3	222	11.4±2.1	186
	$3x$	16.6±2.5	232	8.4±1.4	232
	$2x$	14.0±1.8	238	5.4±1.1	268
Chinese cabbage	$4x$	21.8±1.7	165	10.3±1.7	204
	$3x$	16.6±1.4	195	8.7±1.6	223
	$2x$	14.9±1.7	183	6.9±1.3	268

dividual grown up from the exceptionally small seeds in both the species took an intermediate size between those of the $4x$ and $2x$ ones, directly revealing that the individual is of $3x$ in nature. The number of chloroplasts contained in each cell showed a quite similar mode of variation as those of the cell size. The theoretical ratio of the diameters of those three kinds of the cells of different nuclear contents is calculated to be $\sqrt[3]{2a} : \sqrt[3]{3a} : \sqrt[3]{4a} = 1 : 1.14 : 1.26$. And the following actual ratios were obtained respectively; 1 : 1.19 : 1.37 in radish, and 1 : 1.11 : 1.46 in Chinese cabbage.

II. Cytological observations

Meiotic divisions of the PMCs were examined with several individuals of Japanese radish and Chinese cabbage, which had grown up from the seeds collected from the auto-tetraploid plants selected. As stated elsewhere, all the plants grown from the seeds of exceptionally small sizes were ascertained to be of $3x$, chromosomal behaviours of PMCs in these $3x$ individuals are reproduced in the following:[1]

(a) Triploid radish, $2n=27$.

At heterotypic metaphse several tri- and univalent chromosomes

[1] All the micrographs or camera lucida drawings of the chromosomes obtained were not presented here, and the reproduction of those figures was postponed to some future occasions, in which the problems on the variation or alteration of chromosome association during the years of repeated reproduction in the auto-polyploid forms will be thoroughly treated.

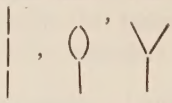
appeared always along with the bivalent ones. Tri- and bivalent chromosomes took regular orientation on the equatorial plane, but univalents were scattered outside of the plate or dispersed on the surface of the spindle sphere. Most III-chromosomes took either one of the following three kinds of shapes at I-Metaphase;  . That of  shape occurred very rarely. No IV-chromosomes was encountered at all. Frequencies of various configurations of the chromosome associations are reproduced in Table 2. The somatic chromosome

Table 2. Chromosome associations at I-Metaphase in $3x$ Japanese radish.

Configuration	Frequency
9_{III}	21
$8_{III}+1_{II}+1_{I}$	41
$7_{III}+2_{II}+2_{I}$	35
$6_{III}+3_{II}+3_{I}$	21
$5_{III}+4_{II}+4_{I}$	4
$4_{III}+5_{II}+5_{I}$	3
Total number of PMCs	125

number of these individuals was exactly determined as 27, i.e., three times the basic set of 9. As shown in Table 2 each nucleus contained 7.35 III-chromosomes in average. From the mode of chromosome associations it is quite certain that those individuals observed would be of the auto-triploid in nature.

Table 3. Frequency occurrences of lagging chromosomes and chromosomal bridges at I-Anaphase in $3x$ Japanese radish.

I-Anaphasic figures	Frequency	
with no laggard	89	59.0 %
with 1 laggard	39	38.4 %
〃 2 laggards	14	
〃 3 〃	4	
〃 4 〃	1	
with 1 bridge	3	2.6 %
〃 2 bridges	1	
Total number of PMCs	151	100 %

At I-Anaphase there appeared some laggards lying behind in the equatorial region, which are duly ascribed as a part of univalent at I-Metaphase. The tri- and bivalent chromosomes disjoined and proceeded to either one of the poles, and some univalents situated in the neighbourhood of the poles went intact to the respective poles with the disjoined chromosomes. And the rest of the univalents situated rather remotely from the equatorial region, remained as laggards between the poles, also showing, in turn, some movement towards the equator and their splitting. In some rare cases, a chromosomal bridge, which discloses the occurrence of some inversions, was encountered at I-Anaphase. Frequency occurrences of those lagging univalents and chromosomal bridges are presented in Table 3. Tri-polar spindles were also met with in some rare occasions.

As the results of uneven distribution of chromosomes at I-Anaphase, the number of chromosomes constituting the homotypic plates showed marked variation between 9 and 18, 13 or 14 being the most frequent. In or out of the equatorial plate consisting of the diad chromosomes there appeared a few monad ones, each of which was considered to be the split half of the univalent. Table 4 is compiled by the data of frequency distribution of chromosomes on each homotypic plate, which was composed exclusively of the diad chromosomes. As may be duly expected, there occurred also some lagging chromosomes at II-Anaphase, and these situations will be, again, ascribed as the cause of abnormal polyads appearing at sproad stage.

Table 4. Frequency distributions of diad chromosomes at II-Metaphasic plates in $3x$ Japanese radish.

Number of diad chromosomes in each II-plate	9	10	11	12	13	14	15	16	17	18	Total number of II-plates
Frequency	1	1	8	23	40	32	22	5	1	1	134

(b) Triploid Chinese cabbage, $2n=30$.

As the basic chromosome set in Chinese cabbage is 10, the autotriploid individuals are consist of 30 chromosomes. A quite similar association procedure of chromosomes as in $3x$ radish was met with at I-Metaphase of microsporogenesis (See Table 5). The average number of III-chromosomes appearing in each nucleus was 8.42. Behaviours of chromosomes in both the first and the second divisions were quite similar to those in $3x$ radish plants. Table 6 shows the lagging chromosomes appearing at I-Anaphase. Table 7 represents the frequency occurrence of monad chromosomes which are situated in the vicinity of sister plates at II-Metaphase. No I-Anaphasic bridge was encountered at all.

Table 5. Chromosome associations at I-Metaphase in 3x Chinese cabbage.

Configuration	Frequency
10_{III}	18
$9_{III}+1_{II}+1_{I}$	29
$8_{III}+2_{II}+2_{I}$	23
$7_{III}+3_{II}+3_{I}$	14
$6_{III}+4_{II}+4_{I}$	5
$5_{III}+5_{II}+5_{I}$	1
Total number of PMCs	90

Table 6. Frequency occurrences of lagging chromosomes at I-Anaphase in 3x Chinese cabbage.

I-Anaphasic figures	Frequency	
with no laggard	28	35.4 %
with 1 laggard	26	64.6 %
〃 2 laggards	15	
〃 3 〃	8	
〃 4 〃	2	
Total number of PMCs	79	100 %

Table 7. Frequency occurrences of II-Metaphasic plates with or without monad chromosomes in 3x Chinese cabbage. Countings were obtained with each set of sister plates.

Number of monad chromosomes appeared in		Frequency	
one plate	another plate		
0	0	68	52.7 %
1	1	25	47.3 %
0	2	18	
2	2	9	
1	3	8	
3	3	1	
Total number of PMCs		129	100 %

Homotypic plates, which consisted exclusively of the diad chromosomes and did not contain any monad ones, showed marked variation as presented in Table 8. Plates consisted of 10 or 20 chromosomes,

Table 8. Distributions of chromosomes at II-Metaphasic plates in 3*x* Chinese cabbage.

Number of chromosomes in each II-plate	11	12	13	14	15	16	17	18	19	Total number of plates
Frequency	2	3	17	17	27	21	12	2	1	102

i.e., the extreme numbers were not obtained, and the plates of 15 occurred most frequently. Several lagging monad chromosomes were also observed at II-Anaphase (See Table 9). In each 3*x* mother-cell there appeared 1.58 univalents in average at I-Metaphase (From Table 5), and 1.11 laggards also at I-Anaphase (From Table 6). Monad chromosomes derived through splitting of diads had amounted to 0.67 per cell in average (From Table 7). From the data calculated above it may be safely deduced that the 70 per cent of the univalents appeared at I-Metaphase remained as laggards on the I-Anaphasic spindles, and that the 56 per cent of the latter, again, have accomplished their splitting, while the remaining ones have proceeded intact towards either one of the poles. Furthermore, Table 7 shows that the monad chromosomes appearing in each II-Metaphasic nucleus were 1.24; and Table 9 reveals that the laggards appearing at II-Anaphase amounted to 1.03 per cell. So that the 80 per cent of the monad chromosomes occurring at I-Anaphase might have appeared again as the laggards at II-Anaphase. There is need to mention here, in addition, that some of the lagging chromosomes would have been excluded from the division spheres, and some of them contributed to the formation of supernumerary microspores.

Table 9. Lagging chromosomes apparent at II-Anaphase in 3*x* Chinese cabbage. Countings were made with each set of homotypic spindles in a PMC.

Number of laggards appeared on one spindle	Number of laggards appeared on another spindle	Frequency
0	0	50
0	1	11
0	2	11
1	1	11
1	2	6
2	2	5
1	4	1
Total number of PMCs		95

All the plants grown up from the seeds of larger sizes, which were produced on the auto-tetraploid individuals, disclosed that they could be divided into two different categories, the former of which was of euploid, and the later was of certain heteroploid, i.e., hyper- and hypo-tetraploid. Results of cytological examinations with those plants are presented in the following:

(c) Tetraploid radishes.

(i) Eu-tetraploid plants, $2n=36$.

Two plants, Nos. 10 and 11, were determined to contain 36 chromosomes in somatic state, revealing that they are of eutetraploid. Reduction divisions of the PMCs in these plants proceeded quite regularly, forming exclusively IV- and II- chromosomes at I-Metaphase (See Fukushima, Tokumasu, and Oguro, 1949). Any other multivalent chromosomes were not encountered at all. But the univalent chromosomes were noticed in some rare cases. Table 10 represents the frequency

Table 10. Frequency occurrences of IV chromosomes at I-Metaphase in $4x$ Japanese radish.

Number of IV-chromosomes in each nucleus		9_{IV}	8_{IV}	7_{IV}	6_{IV}	5_{IV}	4_{IV}	3_{IV}	2_{IV}	1_{IV}	0_{IV}	Total number of PMCs
Frequency	Plant No. 10				5	9	8	14	8	4	2	50
	Plant No. 11	1	1	3	10	9	16	11	8	3	1	63

appearances of the IV-chromosomes per nucleus in Plant Nos. 10 and 11 respectively. Heterotypic anaphase proceeded quite regularly in those plants. In Plant No. 11, however, only one laggard appeared not infrequently. Two or more laggards did not appear in any one cell at I-Anaphase. Homotypic nuclear division progressed also in a regular manner, excepting the occurrence of plates containing one or two chromosomes oriented outside of the plates. So the homotypic laggards were met with at some rare occasions. The number of chromosomes oriented on each II-plate is shown in Table 11. In Plant No. 10 chromosomal bridges were also encountered at I- and II-Anaphases.

Table 11. Frequency occurrences of chromosomes arranged on each II-Metaphasic plate in $4x$ Japanese radish.

Number of chromosomes in each II-plate		16	17	18	19	20	Total number of PMCs
Frequency	Plant No. 10		15	80	16		111
	Plant No. 11	2	17	109	17	1	146

(ii) Hypo-tetraploid plants, $2n=34$.

Two plants, Nos. 6 and 21, were ascertained to be of hypo-tetraploid, containing 34 chromosomes in somatic state. Association procedures of I-Metaphasic chromosomes were $(7\sim2)_{IV}+(2\sim0)_{III}+(12\sim0)_{II}+(2\sim0)_{I}$ in Plant No. 6, and $(6\sim0)_{IV}+(2\sim0)_{III}+(15\sim2)_{II}+(2\sim0)_{I}$ in Plant No. 21. From these facts the two missing chromosomes in both plants were duly inferred to be non-homologous to each other. At I-Anaphase one or two univalents appeared sometimes as laggards, and these univalents did not, in general, split to their halves and remained intact throughout the I-division, and some of them were excluded out of the division sphere. In Plant No. 6 the latter univalents were observed not infrequently, i.e., in the 11.6 per cent of cells examined. Tripolar spindles were also found in some rare cases. Table 12 show the frequency occurrences of the number of chromosomes arranged on the sister plates of II-Metaphase. Division processes after the II-Anaphase stage proceeded quite regularly, resulting in the formation of normal sporads.

Table 12. Chromosomes distributed to each II-Metaphasic plate in hypo-tetraploid, $4x-2$, Japanese radish.

Number of chromosomes in each II-plate		15	16	17	18	19	20	Total number of plates
Frequency	Plant No. 6	11	16	50	13	8	1	99
	Plant No. 21	7	19	32	18	2		78

(iii) Hyper-tetraploid plant, $2n=37$.

Plant No. 20 had contained 37 chromosomes in somatic state, consisting of the tetraploid number of chromosomes and an extra one in addition. Chromosome associations at I-Metaphase showed the configuration, $(7\sim0)_{IV}+(1\sim0)_{III}+(18\sim3)_{II}+(1\sim0)_{I}$. One pentavalent chromosome was, however, observed on some rare occasions. One or two lagging chromosomes appeared at II-Anaphase. Diad chromosomes distributed on the II-Metaphasic plates showed the variation depicted in Table 13. A few monad chromosomes usually occurred at II-Metaphase, though such plates were excluded from the compilation of the table.

Table 13. Chromosome distributed to each II-Metaphasic plate in hyper-tetraploid, $4x+1$, Japanese radish.

Number of chromosomes in each II-plate	16	17	18	19	20	21	Total number of plates
Frequency	2	4	21	27	4	1	59

(d) Tetraploid Chinese cabbage.

(i) Eu-tetraploid plants, $2n=40$.

Among the individuals grown up from the seeds of larger sizes, as stated elsewhere, two plants, Nos. 1 and 2, were ascertained to be of eu-tetraploid. In most I-Metaphasic nuclei there appeared IV- and II-chromosomes, and the exceptional occurrence of a few I- and III-chromosomes was noticed in Plant No. 1. Table 14 shows the number

Table 14. Frequency occurrences of IV-chromosomes at I-Metaphase in $4x$ Chinese cabbage.

Number of IV-chromosomes in each nucleus		8_{IV}	7_{IV}	6_{IV}	5_{IV}	4_{IV}	3_{IV}	2_{IV}	1_{IV}	Total number of PMCs
Frequency	Plant No. 1	1	2	3	6	4	3	4	1	24
	Plant No. 2	3	6	11	11	11	5	5	1	53

of IV-chromosomes occurring in each I-Metaphasic nucleus, their average number amounting to 4.33 and 4.83 in Plant Nos. 1 and 2 respectively. Lagging chromosomes at I-Anaphase were observed to be rather usual. Chromosomal bridges were noticed rarely in Plant No. 2 (See Table 15). The future destiny of these laggards was quite similar to

Table 15. Occurrences of lagging chromosomes and chromosomal bridges at I-Anaphase in $4x$ Chinese cabbage.

PMCs	Frequency	
	Plant No. 1	Plant No. 2
with no laggard	83	82
〃 1 laggard	14	9
〃 2 laggards	2	
〃 1 bridge		2
Total number of PMCs	99	93

Table 16. Frequency of chromosomes composing each II-Metaphasic plate in $4x$ Chinese cabbage.

Number of chromosomes in each II-plate		18	19	20	21	22	Total number of PMCs
Frequency	Plant No. 1	1	13	58	10	1	83
	Plant No. 2		12	82	15		109

that of the laggards described above. Accompanying Tables 16 and 17 will suffice to give a clear insight into the division processes in these eu-tetraploid individuals. Some irregularities, such as the cell with

three homotypic spindles, or with the homotypic bridges, were also met with in a few exceptional cases.

(ii) Hyper-tetraploid plant, $2n=43$.

A Chinese cabbage, Plant No. 3, had contained 43 chromosomes in somatic state, the number being $4x+3$. Chromosome conjugations at I-Metaphase had shown the configuration; $(8\sim0)_{IV}+(3\sim0)_{III}+(18\sim3)_{II}+(2\sim0)_{I}$. Only in some exceptional cases one pentavalent was encountered. From the mode of chromosome associations it would be duly appreciated that the three extra chromosomes added may be non-homologous among each other. From one to three chromosomes lagging behind were observed at I-Anaphase. As shown in Table 18,

Table 17. Lagging chromosomes apparent at II-Anaphase in $4x$ Chinese cabbage. Countings were made with each set of homotypic spindles in a PMC.

Number of laggards in		Frequency	
one spindle	another spindle	Plant No. 1	Plant No. 2
0	0	28	84
0	1	2	11
1	1		1
0	2		1
Total number of PMCs		30	97

Table 18. Frequency occurrences of chromosomes composing each II-Metaphasic plate in hyper-tetraploid, $4x+3$, Chinese cabbage.

Number of chromosomes in each II-plate	19	20	21	22	23	24	Total number of PMCs
Frequency	8	17	50	31	7	2	115

II-Metaphasic plates consisted of from 19 to 24 chromosomes. One or more supernumerary II-spindles were observed not infrequently. A mothercell was encountered, containing a large II-plate of 43 chromosomes, which may have been derived through the restitution phenomenon. At II-Anaphase some lagging chromosomes occurred usually.

III. Fertility

Microspores at sporad stage showed some irregularities in all the polyploid individuals examined. The types and frequencies of those abnormal sporads are shown in Table 19. The hypo-tetraploid radish, Plant No. 6, $2n=34$, and the hyper-tetraploid Chinese cabbage, Plant No. 3, $2n=43$, showed the most prominent abnormality, the extents of

Table 19. Frequency occurrences of various types of sporads formed by polyploid and aneuploid individuals in Japanese radish and Chinese cabbage.

	Polyploidy	Types of sporads*									Total number of sporads	% of sporads other than ④
		①	②	③	④	⑤	⑥	⑦	⑧	⑨		
Frequency — Japanese radish	$3x$	1	–	–	850	6	2	–	–	–	859	1.0
	$4x$ Plant No. 10	1	–	–	361	2	–	–	–	–	364	0.8
	$4x$ Plant No. 11	–	–	–	520	3	–	–	–	–	523	0.6
	$4x-2$ Plant No. 6	–	–	–	351	90	95	18	3	1	558	37.1
	$4x-2$ Plant No. 21	1	2	–	348	2	–	–	–	–	353	1.4
	$4x+1$	–	–	–	750	–	–	–	–	–	750	0.0
Chinese cabbage	$3x$	–	–	–	482	29	2	–	–	–	513	6.0
	$4x$ Plant No. 1	–	–	–	516	16	3	–	–	–	535	3.6
	$4x$ Plant No. 2	–	–	–	365	5	1	–	–	–	371	1.6
	$4x+3$	–	1	–	328	42	3	2	–	–	376	12.8

N.B. * Type ④ denotes the normal tetrad; type ⑤ or ⑦ the pentad or heptad; and type ① the monad.

which far exceeded the expectations derived from the highly irregular behaviours of their meiotic chromosomes. Namely, in those plants pentads or still higher polyads were produced much more frequently than the estimations due to the results of karyological observations. The causes of such a situation is yet, however, not quite certain to the authors. The $3x$ individuals had shown, in turn, only a slight amount of abnormalities in the formation of their sporads, though each tetrad consisted of four spores, taking various sizes and markedly differing in each nuclear content. Thus again, the situation seems not to concur with the expectations deduced from the chromosomal behaviours.

Matured pollen grains were stained with the aceto-carmine, and the morphologically normal and stainable grains were identified as the viable ones. From Table 20, it is clear that the $3x$ plants of both the radish and Chinese cabbage have shown somewhat low degrees of pollen viability, but the percentage values were remarkably higher than those of the auto-triploid plants in various other plant species. Thus the triploidy accompanying such a comparatively high pollen viability seems to be confined to the special characteristics of *Brassica* and *Raphanus*. The radish hypo-tetraploid, Plant No. 6 ($4x-2$), and the hyper-tetraploid Chinese cabbage, Plant No. 3 ($4x+3$), both of which had produced remarkably higher percentages of abnormal polyads, showed clearly, in

Table 20. Morphological viability of pollen grains of polyploid and aneuploid individuals in Japanese radish and Chinese cabbage.

	Chromosomal constitution	Number of pollen grains examined	Number of viable pollen grains*	% of viable pollen grains
Japanese radish	$2x$	1377	1265	91.9
	$3x$	1553	1266	81.5
	$4x$ Plant No. 10	655	645	98.5
	$4x$ Plant No. 11	1064	1022	96.1
	$4x-2$ Plant No. 6	2426	2019	83.2
	$4x-2$ Plant No. 21	776	749	96.5
	$4x+1$	1385	1335	96.4
Chinese cabbage	$2x$	598	592	99.0
	$3x$	1818	1597	86.9
	$4x$, Plant No. 2	1099	1076	97.9
	$4x+3$	769	711	92.5

N.B. * Viability of pollen grains was only inferred from their outward morphology and the staining reaction to aceto-carmine of their plasmatic inclusions.

concurrence with the above situation, slightly lower values of pollen viability than those values in the other tetraploid members. Among the other members of the present tetraploid strains, however, there could not be noticed any clear differences in pollen fertility.

Diameters of the socalled viable pollen grains were measured and compared (See Table 21). Diameters of the grains produced on the $3x$ individuals showed always much greater variability than those of $4x$ or $2x$ ones, though the average diameter took the intermediate value of the latter two. The above circumstances were considered to concur well with those of the meiotic procedures stated elsewhere. Any clear differences in the sizes of the viable pollen grains could not be obtained among the various tetraploidal forms treated above, so it may be ascertained that such a slight variation in the gametic content of chromosomes, extending only to the addition or subtraction of one or two chromosomes, could not be reflected so much on the size of pollen grains.

All the polyploid individuals, including some aneuploid ones, showed rather good setting of the pods after blooming, not showing any noticeable differences among the sister individuals. Owing to the adverse conditions of growing, the exact nature of which could not be made certain, the seed development in Chinese cabbage was very poor, so the

Table 21. Comparisons of diameters of pollen grains in polyploid and aneuploid individuals in Japanese radish and Chinese cabbage.

	Chromosome constitution	Classes of diameters of pollen grains (micrometer unit) 15	16	17	18	19	20	21	22	23	24	25	26	Total number of pollen grains	Mean diameter
Japanese radish	$2x$	10	121	134	197	38	9	1						510	17.32
	$3x$	6	8	15	45	67	25	28	18	11	1		1	225	19.30
	$4x$				1	11	101	126	110	12	1			362	21.03
	$4x-2$				11	41	98	131	106	25	11	1		424	20.95
	$4x+1$				2	22	91	112	105	36	13	1		382	21.21
Chinese cabbage	$2x$	25	58	56	50	21	1							211	16.94
	$3x$			5	21	57	129	126	72	34	9		1	454	20.64
	$4x$				2	7	26	59	70	54	33	21	5	277	22.22
	$4x+3$				1	11	23	52	52	36	23	12	3	213	21.98

data of their seed fertility were discarded. The radish strains, in turn, had provided very reliable data of seed fertility as shown in Table 22. It is worthy of note that the $3x$ individuals of radish produced a fairly large number of good seeds, well competing with those of the $2x$ or $4x$ individuals. Furthermore, the authors could not obtain certain clear differences in seed fertility among the eu-tetraploid and hyper-tetraploid radishes treated in the present studies.

Table 22. Comparisons of seed-fertility in polyploid and aneuploid individuals in Japanese radish.

Chromosome constitution	Number of pods produced	Number of viable seeds	Number of non-viable seeds	Viable seeds/pod	Non-viable seeds/pod
$2x$	429	992	50	2.31	0.12
$3x$	1172	1808	237	1.54	0.20
$4x$	397	697	9	1.76	0.02
$4x-2$	314	550	50	1.75	0.16
$4x+1$	350	603	34	1.72	0.10

Considerations

In the offspring of the autotetraploid plants raised artificially, it is generally experienced that there usually appear the hyper- and hypo-tetraploid individuals, together with the eu-tetraploid ones. Aneuploidy

encountered in the offspring of the present tetraploid strains of *Raphanus* and *Brassica* is a quite similar example. Since the chromosome numbers of the mother plants had not been ascertained beforehand, the exact origin of those aneuploid individuals is not quite certain, but the results of cytological observations with the eu-tetraploid individuals, which have been produced along with those aneuploid ones in the offspring, will suffice to make the situation clear. Uneven distribution of chromosomes at I-Anaphase has occurred not infrequently. As shown in Tables 11 and 12, even from 25 to 30 per cent of the II-Metaphasic plates observed consisted of the chromosomes, whose number was more or less the exact diploid number. The similar phenomenon was also reported by other workers in some species of *Brassica*. Howard (1939 a) reported that in the autotetraploid *B. oleracea* L. there appeared the II-Metaphasic plates consisting of from 16 to 20 chromosomes, the regular plate of 18 amounting to 57 per cent of the plates examined. Yakuwa (1944) also obtained the similar data from his autotetraploided *B. chinensis*. L. Chromosomes constituting the homotypic plates varied between 17 and 22, and the frequency of the regular plate of 20 chromosomes was 64.4 per cent. If the homotypic plates consisting of the unbalanced chromosomes, which have been formed through the uneven chromosome distribution, are capable of growing up into the mature viable gametes, the hyper- or hypo-tetraploid zygotes will naturally result.

It is generally appreciated that the sterility phenomenon accompanying the autopolyploidy would be mainly derived through the meiotic irregularities caused by the occurrence of multivalent associations of homologous chromosomes. As mentioned previously, the eutetraploid individuals of both the radish and Chinese cabbage did not show any great irregularities in their meiotic processes. At heterotypic metaphase, the most chromosomes appeared in IV or II, and III- or I-chromosomes occurred with much less frequencies. Some of those IV-chromosomes, however, were observed to disjoin non-disjunctionally or unevenly at I-Anaphase, and thus a certain percentage of the II-plates with unbalanced chromosomes were formed. In general, the gametes grown up from those nuclei containing unbalanced chromosomes are considered to realize more or less lethal effects upon the reproductive processes. The auto-tetraploid individuals of both the radish and Chinese cabbage produced the II-plates consisting of an exactly balanced number of chromosomes, i.e., 18 or 20 respectively, in 70—75 per cent of the II-plates examined, while the percentages of the viable pollen grains produced by those individuals have amounted to as high as 96—98. Thus in the present eutetraploid individuals it may be duly suggested that the microspores containing unbalanced nuclei can grow up into the matured grains which are supposed to be effective on the fertilization.

As shown in Table 22, the seed fertility of the eutetraploid radishes is somewhat inferior to that of normal diploid ones. And such differences in seed fertility can not be simply attributed to the higher lethality of the zygotes set on the tetraploid individuals, because the percentage occurrence of the nonviable seeds in the tetraploid pods is certainly less than that of the normal diploid ones. Thus the low fertility will be caused by the following alternative: that the eutetraploid individuals may produce the pods, containing a definitely smaller number of placentæ than that of diploid, or that the diploid gametes produced may have decisively lower reproductive power than the normal haploid ones.

In both the Japanese radish and Chinese cabbage, the gametes with unbalanced chromosomes could develop almost regularly and effect the fertilization, producing viable seeds. Such a circumstance is most outstanding on the present autotriploid forms, because almost all the gametes produced are containing the nuclei of more or less unbalanced nature. Table 20 indicates that the percentages of the morphologically viable pollen grains in those $3x$ radish and Chinese cabbage plants were 82 and 87 respectively. And moreover, the $3x$ radish produced almost as many good seeds as those of the $4x$ individuals (See Table 22). The greater variation ranges of the diameter readings prevailing on the matured pollen grains in the $3x$ individuals is considered to reflect, no doubt, the situation in the nuclear contents. It thus appears that the $3x$ individuals can reproduce under self-fertilization various kinds of individuals, whose chromosomal constitutions varied markedly between the $2x$ and $4x$ numbers.

Occurrences of the hyper- or hypotetraploid individuals in the offspring of the present autotetraploid strains would be, in turn, attributable to the conjugation of the viable gametes having the aneuploid number of chromosomes. Aneuploid individuals thus resulting, having one or two chromosomes in addition or in subtraction to the eutetraploid number, did not show any discernible morphological differences from the eutetraploid individuals in the same offspring. And certain irregularities in the meiotic processes, accompanying the aneuploidy, did not, however, lead to the diminution, even to a very slight extent, of the apparent pollen viability and also of the actual seed fertility. Such circumstances may have been derived from the fact that there were no clear differences on the effectiveness to fecundation between the two categories of gametes, one consisting of the euploid number of the chromosomes and the other of the heteroploid ones. And furthermore, size of matured pollen grains developed from the heteroploid microspores, so far as the heteroploidy of the present cases is concerned, could not realize any discernible distinctions from the ordinary euploid ones. Thus it seems very interesting to infer that those aneuploid

individuals were almost indistinguishable from the sister euploid ones in the several characteristics depicted as follows: the general morphological appearances of the plant status; the percentage of pods set per flowers; the sizes and the viability of the matured pollen grains; and the actual seed fertility.[1]

As mentioned previously, it has been experienced usually that the artificially raised tetraploid strains return unawares to the original diploid ones under cultivation through generations, being deprived of the polyploid nature of nuclei. The causes of such regressional tendencies in autopolyploidy are duly considered by the present authors to be attributable, in the main, to the usual occurrence of aneuploid individuals in the offspring, because once the diminution of the chromosomes has commenced it will progress steadily and rather rapidly towards the final diploid state. As mentioned elsewhere, the present tetraploid strains of both the radish and Chinese cabbage have already passed through several generations and have been maintained as such with some stability. But, as was presented in the authors' results, the individuals with an exact tetraploid number of chromosomes had still produced a certain number of tetravalent chromosomes and developed many viable gametes consisting of various aneuploid numbers of chromosomes. In consequence, when an original polyploid plant is once induced artificially, its descendants will henceforth continue to separate out the aneuploid individuals in a certain progressive ratio through successive generations. And then, if the seed gatherings were intended to be made under self-pollinations from only a very few member in a definite strain, for an experimental or a practical culture, there may always accompany the opportunities for selecting the undesirable aneuploid individuals, and thus the heteroploidal tendencies in much stronger degrees may come to prevail over the future generations, providing finally the diploid condition. And moreover, it is not very easy to avoid such circumstance since it is quite difficult to identify the exact nature of those aneuploid individuals from the euploid ones throughout their whole vegetative stages and even at their blooming time. To maintain a certain tetraploid strain, we are accustomed, in general, to make the examinations upon the size and stainability of matured pollen grains with each individual. But as stated elsewhere, the size of the pollen grains of both the radish and Chinese cabbage does not reveal its gametic nature exactly, so that the actual chromosome counting

[1] The characters and behaviours of the present aneuploid individuals may be taken as rather exceptional ones, and the noteworthy case of the offspring of the colchicine-induced eutetraploids of *Antirrhinum majus* (Straub, 1941) seems to be one of the typical examples. Even the aneuploids such as $4x+1$ or $4x-1$ forms could be easily identified through their several morphological characters and their definitely low pollen viabilities.

becomes a necessary task to make clear the genetic status of each individual. For the practical seed collecting with the tetraploid strains, the counting of chromosomes with each plant seems, however, to be near to improvable measures, and thus the more practicable task of maintaining the polyploid status is to collect seeds from as many individuals as possible. And further, it must be taken into consideration that the seeds of $3x$ constitution are quite small in size and are identifiable very easily. Howard (1939b, 1942) reported that the $3x$ seeds of *B. oleracea* and of *B. chinensis* are very small in size, taking about one third of the ordinary size, but filling fully the testa. Nishiyama (1949b) has also noticed that the $3x$ seeds of *Raphanus* were similarly small in size, and that he could easily sift out such small ones from all the other seeds produced on the $4x$ forms. As described elsewhere, with both of the present $3x$ radish and Chinese cabbage a quite similar situation in seed size was ascertained by the authors.[1]

It is well inferred that the tetraploid strains grown in the vicinity of the diploid relatives will easily produce such $3x$ seeds by the natural crossings. The authors noticed that the frequency occurrence of those $3x$ seeds had varied more remarkably among the individuals belonging to the same $4x$ strain than among those belonging to the different varieties or strains. Nishiyama (1949b) has also reported that the natural crossability between $4x$ and $2x$ individuals of the same variety decreased according to the increase of planting distance between $4x$ and $2x$ forms, and it showed again marked variations among the different combinations of varieties undertaken in the adjacent planting. Thus it may be safely concluded that even among the tetraploid individuals of the same strain there may exist rather definite differences on the crossability with the pollen grains from the same diploid source. So far as the present studies have gone, the causes of such a situation are not yet made clear, although some environmental and genetic causes may well be inferred.

During the procedures of chromosome diminution towards the diploid state in the tetraploid offspring through the aneuploidy, a certain amount of seeds in extremely unbalanced state of nucleus, such as those in the triploid or its neighbourhood, will, no doubt, result. And the latter seeds are duly supposed to have very small sizes. To the

[1] In connection with such a situation, the authors remember the fact that the various interspecific crosses in *Brassica*s, covering the diploid, tetraploid, and the amphidiploid species or strains raised artificially, there usually occurred true F_1 hybrid seeds of very small size; or, in other words, that the true F_1 seeds could only be gathered among the seeds of extremely small sizes produced on the crossings (See Howard (1942), Hosoda (1949), and Iwasa (1951)). It seems rather an easy task to exclude those small hybrid seeds occurring spontaneously from the seeds to be collected for maintaining a definite polyploid strain.

authors, the exact relationship between the smallness of size of seeds and their unbalanced states of chromosome construction is not yet certain,[1] but the selection of seeds by their sizes will be certainly taken as one of the practical measures for maintaining a certain tetraploid strain of vegetables, such as Japanese radishes or some *Brassica*s.

Summary

1. To maintain the chromosome constitution of certain newly raised auto-tetraploid forms through generations is one of the urgent claims in plant-breeding by the chromosome doubling, because it has been generally experienced that the auto-tetraploid forms are apt to return always to the former diploid state even under the usual seed-growing measures. Some cytological evidences concerning such procedure of chromosome diminution are presented.

2. The auto-tetraploid individuals of both Japanese radish and Chinese cabbage produced a large number of $4x$ seeds of ordinary size together with a fairly large number of $3x$ seeds, which are quite full but very small in size, taking almost one third of the $2x$ size. Triploid seeds are considered to be due to the natural crossings between $4x$ and the pollens from diploid sources. Diploid seeds took, in turn, a somewhat intermediate size between the $4x$ and $3x$ ones.

3. Comparisons of certain morphological and physiological characters were made with many individuals grown up from those seeds set on the eutetraploid plants. Sizes of cotyledons of $4x$ plants were the largest, those of $2x$ medium, those of $3x$ being the smallest. Triploid individuals showed quite vigorous growing as a whole and their blooming was most delayed and fell behind $4x$ and $2x$ ones. Sizes of

[1] Howard (1942) presented some considerations for the explanations of the occurrence of those small $3x$ seeds in *Brassica*s. He has suggested the application of Watkins' (1932) general rule in crosses of the type high-chromosome number × low-chromosome number that better seed production was obtained with the high-chromosome number one as female and it may be due to the genomic embryo : endosperm relation. And the small size of the $3x$ seeds was explained as being due to an abnormal embryo : endosperm relation, a $3x$ embryo developing in a $5x$ instead of in a 4.5-ploid endosperm (c.f., the normal-sized seeds from both $2x$ selfed and $4x$ selfed which have normal embryo : endosperm ratios of 1 : 1.5). For the explanation of the reduction in seed size in the interspecific crosses he has extended the above idea of embryo : endosperm ratio from the total genomes present to numbers of each type of genome present. Thus in a cross, e.g., *B. chinensis* female × *B. carinata*, 1 *chinensis* genome in the embryo is balanced against 2 instead of 1.5 *chinensis* genomes in the endosperm, and 1 *carinata* genome in the embryo against 1 instead of 1.5 *carinata* genomes in the endosperm. Such a disturbance of normal embryo : endosperm ratios is considered to take place in every interspecific cross.

the stomatal guard-cells and the number of chloroplasts in each guard-cell were measured and compared, revealing the following sequence of decreasing order with both items: 4*x*, 3*x*, and 2*x* plants.

4. Among the offspring of both the 4*x* radish and Chinese cabbage there usually appeared a few hyper-or hypo-tetraploid individuals along with the eutetraploid ones. Occurrence of those aneuploid individuals was suggested as due to the chromosomal behaviours of 4*x* meiosis and the resultant formation of the not less amount of viable gametes consisted of the aneuploid numbers of chromosomes.

5. The formation of such viable and effective aneuploid gametes in both 4*x* radish and Chinese cabbage concurs well with the fact that their 3*x* relatives undertook nearly regular meiotic behaviours and showed, in consequence, rather high pollen and seed fertility, being only slightly lower than those in the euploid forms, 4*x* or 2*x*.

6. Through the above situation of producing the effective aneuploid gametes in some definite ratios to the typical 2*x* gametes, the 4*x* strains are always confronted with contamination of a fraction of heteroploid forms, so that the possibility of picking up undesirable heteroploid seed-plants for the reproduction is not so small that we may be almost free of the diminishing tendency of chromosomes in that strain, which is destined to restore easily the original 2*x* condition without special precautions.

7. In order to maintain a certain polyploid strain, the counting of chromosomes with a large number of individuals is the most desirable task, but that is rather difficult and troublesome in practice. Therefore, it may be duly suggested that we should gather the seeds for propagation from as many seed-plants as possible.

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Horticultural Laboratory,
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Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11, No. 1
August 30, 1957

On the influence of the auxins and the anti-auxin upon vernalization[1]

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Introduction

Many researches have already been made on the relation between the auxins and photoperiodism (Dostál and Hošek (2), Leopold and Thimann (11), Bonner and Thurlow (1), Harder and van Senden (5), Fisher and Loomis (3), etc.), and not a few works have also been published on the relationship of anti-auxin and flower initiation (Zimmermann and Hitchcock (12), Galston (4), Leopold and Thimann (11), Bonner and Thurlow (1), Fisher and Loomis (3), etc.) but concerning the correlation between the low-temperature treatment and the auxin as well as the anti-auxin there are found a comparatively small number of investigation besides those conducted by Leopold (Leopold and Guernsey (8, 9), etc.).

The authors, who took an interest in the study of vernalization, carried out researches on the influence of the auxins and anti-auxin, given at various stages of the low-temperature treatment, upon flower initiation.

Materials and methods

As material, seeds of the "Minowase-Daikon," a race of Japanese radish plant (*Raphanus sativus* L. var. *raphanistroides* Makino), were used. This plant needs almost strictly a low temperature treatment for its flower initiation.

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The seeds were first cleansed with water; then soaked in water, or on some occasions in the auxin or anti-auxin solution, they were left in room temperature or placed in an incubator of 30°C; after 20–30 hours, when the radicles had elongated to be about 2–3 mm long, the seedlings were selected and divided into 2 lots; the first lot was treated with low temperature and the second lot was left in the room temperature or placed in an incubator of 30°C, and during these thermal treatments some parts of each lot were drenched respectively in auxin and in anti-auxin solution and one part in water.

The growth hormones used were α–naphthalene acetic acid (NAA) and potassium salt of β–indole acetic acid (abbreviated: KIA); and the anti-auxin was triiodobenzoic acid (TIBA) (prepared by Ishihara Sangyo Kaisha, Ltd.).

Experiment I

The solutions of NAA and of KIA were prepared in four degrees of concentration, namely 0.1, 0.02, 0.01 and 0.002%, and in each of these

Table 1. Application of NAA and potassium indole acetate to the seedlings during the low-temperature treatment.

Growth substance	Concentration (%)	Days of low-temperature treatment	Number of individuals planted [1]	Bolting percentage: Number of days from the last day of low-temperature treatment to the day of observation		
				23	28	31
Water (control)	–	6	44	0	7	8
		8	46	13	43	64
NAA	0.02	6	14	0	0	0
		8	2	0	0	0
	0.01	6	15	0	0	0
		8	4	0	0	0
	0.002	6	17	0	0	0
		8	13	0	17	22
KIA	0.1	6	17	0	0	0
		8	28	0	15	40
	0.02	6	25	0	5	10
		8	37	5	34	61
	0.01	6	20	0	5	6
		8	31	6	27	50
	0.002	6	20	0	5	6
		8	25	0	17	36

[1] Six-day-treated seedlings were planted on July 26, and 8-day-treated on July 28, 1952.

and in water a part of the seedlings were dipped during the period of low-temperature treatment (Table 1).

At the end of 6-day or 8-day low-temperature treatment they were transplanted into gardens; several weeks later the bolting individuals were counted.

Of the individuals treated 6 days in NAA solutions none bolted, while the controls which were treated in water for 6 days, showed only a small bolting percentage.[1] Those treated 6 days in solutions containing less than 0.02% of KIA bolted at a nearly equal rate to the controls, but those in 0.1% KIA solution did not bolt.

Several bolted among those under the 8-day treatment in water, but the 8-day treatment in NAA solution failed, except with individuals in 0.002% solution, which showed a low bolting percentage. On the other hand individuals treated in KIA solutions showed bolting percentages of 36~61, that is, nearly the same percentages as shown by the controls.

Experiment II

Besides the intact seedlings the materials from which cotyledons were cut off were also examined by the use of 0.01% or 0.05% NAA solution during the low-temperature treatment (Table 2).

Table 2. Application of NAA to the intact seedling as well as the cotyledonless one during the low-temperature treatment.

Concentration of NAA (%)	Preliminary treatment	Days of the low-temperature treatment	Number of individuals planted[1)]	Bolting percentage: Number of days from the last day of low-temperature treatment to the day of observation						
				33	36	39	42	46	52	72
Water	Cotyledons left intact	5	20	0	0	20	40	65	100	100
		8	20	0	35	75	100	100	100	100
0.01	〃	5	40	0	0	43	60	60	70	100
		8	40	0	18	58	70	98	98	100
0.05	〃	5	19	0	5	11	16	47	94	100
		8	14	0	0	0	0	64	100	100
Water	Cotyledons cut off	5	19	0	0	11	11	57	78	100
		8	19	0	0	0	37	68	89	100
0.01	〃	5	14	0	0	0	0	14	50	100
		8	7	0	0	0	0	0	29	100

1) Five-day-treated seedlings were planted on April 8, and 8-day-treated on April 11, 1953.

1 Bolting percentage: "number of individuals bolting or shooting into a flowering stalk" ÷ "total number of the plants under test" × 100.

As to the intact seedlings there was no remarkable difference in the bolting percentage between those treated in 0.01% solution and those in water; but comparing those treated in 0.05% solution with the water-treated, it could be seen that the former was behind in bolting in respect of time.

Cotyledonless seedlings, when treated in 0.01% NAA, likewise required more time for bolting than when treated in water.

On the other hand the cotyledonless seedlings treated in 0.01% NAA fell in time of bolting behind the intact seedlings similarly treated.

Experiment III

Seedlings were immersed in KIA solution during the low- as well as the high-temperature (30°C) treatment (Table 3).

Table 3. Application of potassium indole acetate to the seedlings during the period of low-temperature treatment or during the same lapse of time in an incubator of 30°C.

Duration of the treatment and the temperature	Concentration of the solution (%)	Number of individuals planted[1]	Bolting percentage: Number of days from the last day of the treatment to the day of observation						
			33	36	39	42	46	52	72
5 days 5±2°C	Water	30	2	13	43	57	80	93	100
	0.005	31	0	0	19	59	65	90	100
	0.01	30	0	0	10	30	70	93	100
5 days 30°C	Water	16	0	0	0	0	0	14	43
	0.005	17	0	0	0	0	0	0	29
	0.01	22	0	0	0	0	0	5	27
8 days 5±2°C	Water	35	0	17	63	86	100	100	100
	0.005	33	0	12	55	76	100	100	100
	0.01	27	0	0	37	70	100	100	100
8 days 30°C	Water	29	0	0	0	0	0	7	31
	0.005	31	0	0	0	0	0	20	39
	0.01	23	0	0	0	0	0	4	14

[1] Five-day-treated seedlings were planted on April 8 and 8-day-treated on April 11, 1953.

Of the materials of the 5-day low-temperature treatment, those immersed in KIA solution were somewhat later in bolting than those

in water. As to the materials treated with low-temperature for 8 days, there was scarcely any difference in bolting between those immersed in KIA and those in water; but there was a tendency that the higher the percentage of KIA-solution the lower was the bolting percentage.

The other set, which was held at 30°C during KIA treatment, showed a much lower percentage of bolting, similar to the material which was subjected to the same temperature in water.

As stated above, this plant essentially cannot bolt without low-temperature treatment, but it did bolt in this experiment, though in a very low percentage. The reason might be that as the materials were transplanted into the garden in early April, they were affected by a low temperature of less than 8°C several times, and thus some of the materials were vernalized.

Table 4. Application of TIBA to the seedlings during the low-temperature treatment or during the same period of time in an incubator of 30°C.

Duration of the treatment with the solution and the temperature	Concentration of the solution (%)	Number of individuals planted[1]	Bolting percentage: Number of days from the last day of the treatment to the day of observation						
			33	36	39	42	46	52	72
5 days 5±2°C	Water	86	0	15	23	33	63	83	100
	0.01	36	0	0	6	19	47	89	100
	0.02	34	0	3	29	38	68	94	100
	0.05	40	0	0	15	28	30	85	100
5 days 30°C	Water	34	0	0	0	0	0	0	24
	0.01	35	0	0	0	0	0	0	20
	0.05	46	0	0	0	0	0	0	22
	0.1	52	0	0	0	0	0	0	22
8 days 5±2°C	Water	83	0	16	59	78	94	100	100
	0.01	30	0	13	57	83	87	100	100
	0.05	38	0	0	21	45	92	97	100
	0.1	38	0	3	21	66	90	92	100
8 days 30°C	Water	50	0	0	0	0	0	0	45
	0.01	46	0	0	0	0	0	0	38
	0.05	34	0	0	0	0	0	0	38
	0.1	43	0	0	0	0	0	0	33

[1] Five-day-treated seedlings were planted on April 8 and 8-day-treated on April 11, 1953.

Experiment IV

TIBA solution was used during both the low- and high-temperature treatment (30°C) (Table 4). Materials both for 5-day and for 8-day low-temperature treatment, immersed in TIBA solution, differed very little in bolting percentage respectively from those immersed in water, but there could be seen a tendency that the solution of higher percentages retarded bolting to some extent.

Low percentages of bolting were observed on the 72nd day after the transplantation into the garden in all the lots of materials which had been immersed in TIBA during the 5-day or 8-day high-temperature treatment, as well as in the lots of those immersed in water during the same respective treatments; only the former showing rather lower bolting percentages than the latter. Here also the fact of bolting observed, though scantily, in the plant not artificially subjected to low temperature, might be explained by the same circumstance already mentioned in Experiment III.

Experiment V

Materials were immersed in NAA or in TIBA for ten days, that is, 3 days before plus 7 days during the low-temperature treatment (Table 5). It should not be said that there existed any remarkable

Table 5. Application of NAA or TIBA to the seedlings, prior to as well as during the low-temperature treatment.

Seeds were soaked, prior to the treatment, for 3 days with:	Germinated seeds were treated with low-temperature (6±1°C) for 7 days, drenching in:	Number of individuals planted [1]	Bolting percentage							
			Number of days from plantation to observation							
			21	24	27	30	35	39	45	49
Water	Water	26	0	0	0	0	12	21	41	54
NAA (0.002%)	NAA (0.002%)	27	3	3	3	7	22	31	52	56
TIBA (0.002%)	TIBA (0.002%)	19	0	0	0	4	7	11	14	16
Water	NAA (0.002%)	31	0	0	0	0	6	11	31	61
Water	TIBA (0.002%)	26	0	0	0	6	11	11	19	31

[1] Planted on March 25, 1954.

difference in the bolting percentage between seedlings treated in NAA (0.02%) and those treated in water. Seeds immersed in TIBA (0.02%) showed a lower bolting percentage than those immersed in water. In another case, materials were treated with solutions merely for 7 days,

namely, during the low-temperature treatment only. In this case, seedlings immersed in NAA (0.002%) solution were almost equal in bolting percentage to those in water, while materials drenched in TIBA (0.002%) turned out inferior to those in water. As to the NAA solution treatment, there was scarcely any difference between the material which was subjected to it for a period of ten days including 3 days preceding the 7 days of the low-temperature treatment and the material which underwent it only during the 7 days of the low-temperature treatment. Comparison between the plants immersed in TIBA during the period covering the 3 days prior to and the 7 days of the low-temperature treatment and those immersed only for the period of the low-temperature treatment showed that the former were somewhat lower than the latter in bolting percentage, though not remarkably so.

Experiment VI

In this experimentation a lot of the material which was treated

Table 6. Application of TIBA to the seedlings during as well as after the low-temperature treatment.

Germinated seeds were treated with low temperature for 13 days, being drenched in:	After-treatment	Number of individuals planted	Bolting percentage Number of days from the finish of the low-temperature treatment[1] to the observation					
			28	33	38	43	50	58
Water	Seedlings were trasplanted into gardens immediately after the low-temperature treatment	30	0	3	40	63	100	100
NAA (0.002%)		32	6	16	70	78	100	100
TIBA (0.002%)		25	0	0	0	0	24	58
Water	Seedling were drenched in NAA (0.002%) for 4 days, after the finish of the low-temperature treatment until the transplantation	32	0	6	56	78	97	100
Water	Seedlings were drenched in TIBA (0.002%) for 4 days after the finish of the low-temperature treatment until the transplantation	31	3	13	42	62	97	100

[1] The low-temperature treatment was finished on Oct. 10, 1954.

with TIBA or NAA during the low-temperature treatment of 13 days, was compared with a lot which was similarly treated for 4 days after the close of the low-temperature treatment (Table 6). No difference was noticed between the results of the two lots of material treated with NAA; but concerning the material treated with TIBA, the first mentioned lot exhibited a tendency by far stronger than the other to inhibit flower-initiation.

It may also be said that, as to the material treated with solutions during the low-temperature treatment, the NAA-treated was equal to the water-treated, while the TIBA-treated was far inferior to the water-treated, and that concerning the material treated with solutions after the low-temperature treatment, the bolting percentages were approximately the same whether the material was treated with NAA or TIBA or in water.

Discussion

On a general survey of those experimentations it may be said that the bolting percentage was not increased by KIA or NAA applied prior to, in the course of, or after the low-temperature treatment, excepting the cases where, by the application of NAA (0.002%) prior to or in the course of the treatment as seen in Exper. 5 (Table 5) and of NAA (0.002%) in the course of the treatment as seen in Exper. 6, the bolting percentage was somewhat promoted in terms of time. This, however, should not be stated with certainty as the number of individuals treated was small, whereas it was generally noticed that the higher concentrations of those auxins very likely induced the lower bolting percentages.

Concerning TIBA, it was said that its application after the low-temperature treatment gave an almost equal result with the controls (without TIBA); nevertheless the application prior to or during the low-temperature treatment led to a bolting percentage lower than in the controls; only in the case of Exper. 4 the inhibiting effect was scarcely observed.

In the absence of low-temperature treatment, or rather in a treatment with high-temperature (30°C), the application of NAA showed a very poor bolting percentage, as bad as with the controls (with no NAA application). Leopold and Guernsey (9) succeeded in hastening flower initiation by using the low-temperature treatment and auxin application jointly. In the present experiments, though the degree of concentration of KIA and of NAA, as well as the duration of treatment, was not sufficiently varied to make it reasonable to draw a general conclusion, yet it might be said at least that NAA or KIA application could neither be substituted for the low-temperature treat-

ment nor have a power to strengthen the effect of the treatment as far as the present experimentation was concerned. From the fact that several materials used by Leopold and Gurnsey flowered to some extent without either auxin application or low-temperature treatment, whereas the plant which was used in the present experiments almost completely fails to bolt without low-temperature treatment, it might be assumed that the former were qualitatively different in flowering maturity from the latter.

It was mentioned that in Exp. III and Exp. IV the materials which were not intentionally treated with low-temperature bolted to some extent, but that by nature this plant does not flower unless treated with low temperature; yet in these cases the materials were transplanted into the garden in early April and so were exposed several times to temperatures lower than 8°C, and perhaps it was for this reason that some of them vernalized and bolted.

In the present experiments, the TIBA-application caused some inhibiting effect upon flower initiation. If the auxin-level has a delicate action upon flower-initation as stated by Lang (7), it will be considered that an application of TIBA disordered the balance of auxin-level and inhibited bolting.

The length of the period of low-temperature treatment was 6 or 8 days in this experiment. This material was on the whole vernalized completely by the 8-day treatment, but 6 days were somewhat too short for vernalization, being just the length of time for the incipient stage; if an exogenous application of auxin or anti-auxin had had an influence upon flowering, the most evident result could have been noticed; however, the auxin application showed practically no visible modification; only TIBA induced an inhibitory effect. The effect of TIBA was probably produced at the very time of low-temperature treatment and seemingly not as an after-effect upon the later stage of development of the seedling, because TIBA, given after the end of the low-temperature treatment, indicated no inhibitory effect.

The results of the previous work of Kojima, Yahiro and Inoue (6) which demonstrated the fact that the bolting percentage of cotyledonless plants was lower to some extent led to the comparative experimentation on the intact and the cotyledonless seedlings, in Exp. II. If auxin be made in cotyledons, and by cutting off the cotyledon the amount of auxin be reduced to scarcity or zero, the exogenous application of auxin will be expected to have a positive influence upon flower initiation; yet the experimental result in question did not answer to this expectation.

Résumé

1) The material used was the seed of the Japanese radish plant which in almost all cases fails to flower-initiate, without being subjected to low temperature.

It was examined whether the bolting percentage was changed or not by application of solutions of several grades of concentration of NAA, potassium indole acetate, or TIBA, prior to, in the course of, or after the low temperature treatment.

2) The application of NAA or KIA to young seedlings sometimes lowered the bolting percentage and in other cases showed no difference from the bolting percentage of the seedlings left without application; yet there was scarcely found any case where the application exactly induced an increase in bolting percentage.

3) The exogenous application of NAA to cotyledonless seedlings could not increase the bolting percentage; it showed rather an inhibiting influence upon bolting.

4) In general, materials treated with TIBA were lower in bolting percentage than those without TIBA treatment; but application of TIBA after the close of the low-temperature treatment showed no inhibitory effect.

5) The materials without low-temperature treatment or those subjected to 30°C instead of low temperature, did not bolt within 46 days after the transplantation. After applying KIA or TIBA, those materials still did not improve their bolting percentage.

6) Though it is too hasty to draw general conclusions from the present experimentations, yet it may be said that the application of NAA, KIA or TIBA to seedlings in their very early stages neither had the effect to induce flower initiation of plants which were not subjected to low-temperature, nor to promote the bolting of plants already vernalized.

Thanks are due to the grant from the Science Research Fund, Ministry of Education, out of which the expense for this research was partly defrayed.

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Journal of the Faculty of Agriculture, Kyūshū University, Vol 11, No. 1
August 30, 1957

Physiological and ecological studies of *Digitaria* plants. VII
On the external forms and germination of the seed of the Hispid type of *Digitaria adscendens* (H.B.K.) Henrard

Masamoto Shimizu

Introduction

Kondo and others (1935) reported on the external forms of the seed of *Syntherisma sanguinalis* Dulac var. *cilialis* Honda.

But the detailed reports on the forms of the seed of *Digitaria adscendens* (H.B.K.) Henrard (this name is the synonym of the species above mentioned) are not seen, and the papers concerning the germination process of it are not found, and so the author has studied the Hispid type* of the species.

Observations of the seed forms and its germination process are useful to elucidate the mechanism of dormancy in the seed.

It is my pleasure to record here a debt of gratitude to Professor H. Kojima for his kindness in leading me in this study.

The forms of the seed†

Inflorescence of 5–10 racemes (rarely more or less). The racemes, 4–18 cm long, very slender, are digitate at the apex of the culm, finally spreading.

Spikelets in pairs, along one side of the racemes, contiguous, short-unequally pedicelled, falling entire at maturity, somewhat flattened on the back, oblong, pointed.

* The Hispid type of *Digitaria adscendens* (H.B.K.) Henrard is the same as *D. adscendens* Henrard var. *fimbriata* (Link) Henr. which had been described in a previous paper of the author (1956).

† In this paper spikelet was called "seed."

The spikelets are placed with the back of the lemma against the rachis, that is, with first glume, second glume, third glume, lemma, and palea of the fertile floret outward (Fig. 1).

Spikelets, about 3.5 mm long, about 1.0 mm wide, total weight of 1000 seeds measured immediately after the harvest was 1.3166–1.3232 gm and the air dried weight (weighed at the 394th day after the harvest) was 0.8660–0.8767 gm.

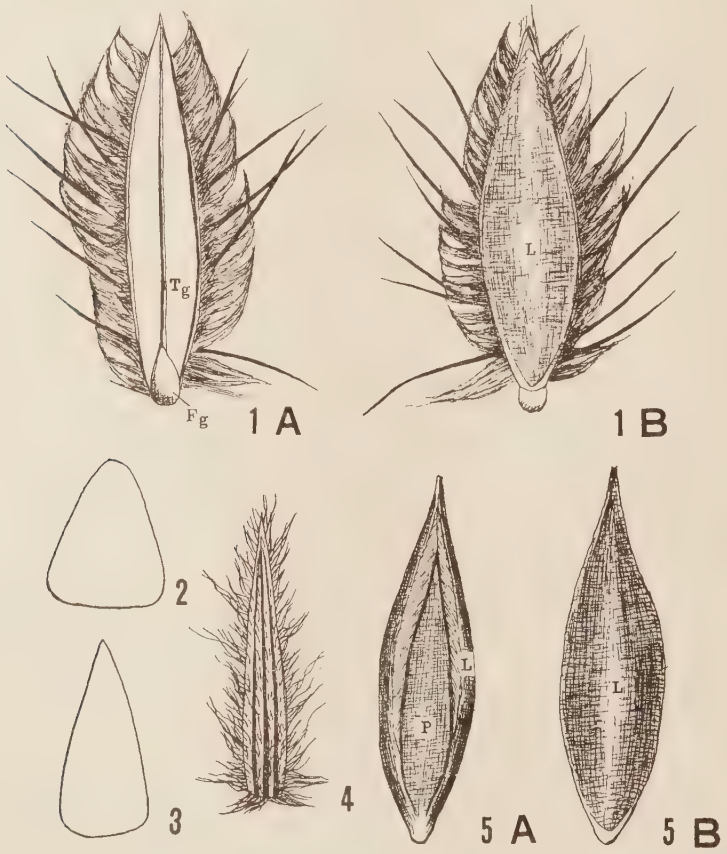

Fig. 1.

1. Two views of a spikelet. ×14.3
   1A: Front view. 1B: Back view, the second glume was removed.
2. The minute palea inside the third glume. ×43.3
3. The First glume. ×43.3
4. The Second glume. ×14.3
5. Two views of a spikelet; glumes were removed. ×14.3
   5A: Front view. 5B: Back view.
   Fg: First glume. L: Lemma. P: Palea. Tg: Third glume.

First glume outermost, minute but evident, triangular, about 0.8 mm long, 0.3 mm wide (Fig. 1, 3).

Second glume is on the opposite side to the first glume, lanceolate, pointed, 3-nerved, thin, 2.0–2.2 mm long (Fig. 1, 4).

Third glume flat or nearly so, widely lanceolate, the length and the width are similar to those of the spikelet without the third glume; strongly 3 nerved, the lateral nerves pubescent, the hairs 0.7–1.05 mm long, and tips of the hairs range in a row, and beside the hairs there

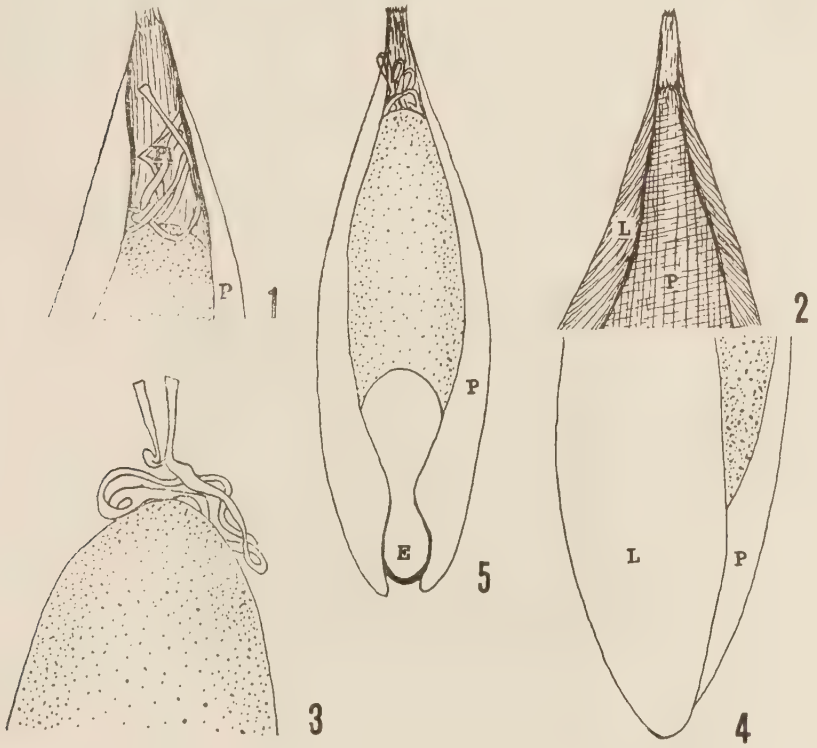

Fig. 2.

1. A pointed end of the palea, showing the shriveled pistil in an open space. ×52.
2. A pointed end of a spikelet; the glume was removed, showing the minute serration. ×52.
3. The shriveled pistil. ×52.
4. The base of a caryopsis enclosed by the hull; the lemma was removed. ×27.2.
5. The caryopsis is enclosed by the palea; the lemma was removed ×27.2.

E: Embryo. L: Lemma. P: Palea. Pi: Shriveled pistil.

exist some 5 brestles pectinately (2.0–2.8 mm long) on the back side of each hair zone (Fig. 1, 1A, 1B).

Third glume has a minute palea inside* (Fig. 1, 2). Lemma, as long as the third glume (and these two decide the length of the spikelet), pointed, firm except for the thin margin, smooth, darkbrown at maturity, oblong, being rounded on the back. Its margins tightly embrace the narrower and shorter inner scale known as the palea. The palea is flattened on the back (Fig. 1, 5).

The pointed tops of the lemma and palea minutely serrate (Fig. 2, 1, 2), and shriveled pistil remains in the open space of the top part (Fig. 2, 1, 5), and the shriveled stigma is often seen from the outside.

The caryopsis of the dormant seed is enclosed so tightly by hulls (lemma and palea) that it can not be stripped readily, but when the seed awakes from its dormancy the caryopsis separates from the hull easily.

Caryopsis 2.2–2.5 mm long, 0.8–0.9 mm wide, (Fig. 3) bears a shield-shaped body, the embryo, yellowish brown on its rounded side at the

Fig. 3. Two views of a caryopsis. ×16.4
Left: Back view, showing embryo.
Right: Front view, showing hilum.

base and the other side of the caryopsis is flattened, with the hilum at the base.

The shriveled pistil on the top of the caryopsis about 2.4 mm long, 10 μ wide; its two stigmas stand in parallel (Fig. 2, 3).

* The spikelet of this genus has one perfect terminal floret and a sterile floret below, so I suppose that the third glume and the minute palea inside it correspond to a rudiment of the sterile floret.

Especially on the visible morphological changes on the embryo during the germination period.

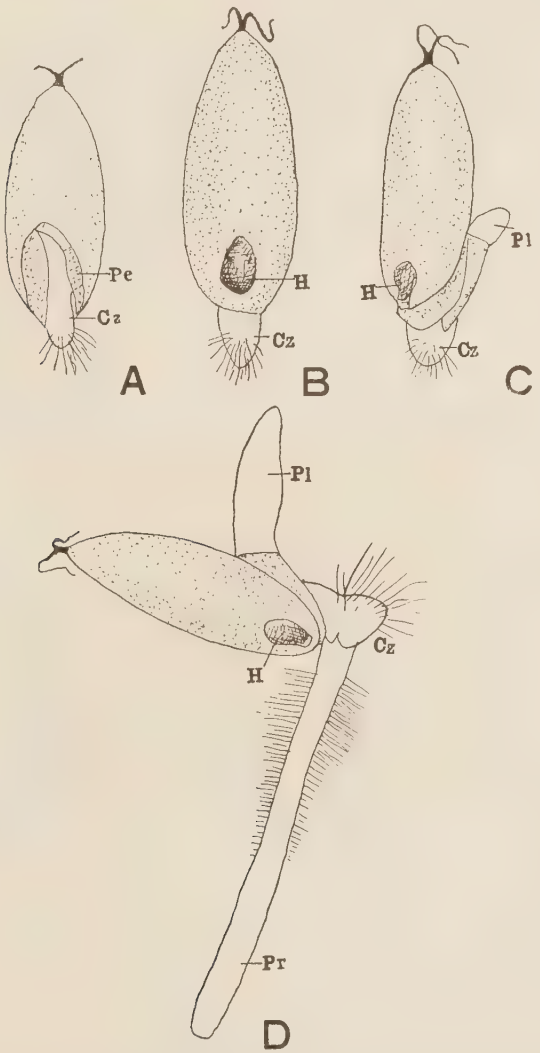

Fig. 4. Germination of the caryopsis, showing rupture of the pericarp by the coleorhiza and growing plumule and primary root. ×16.4
Cz: Coleorhiza. H: Hilum. Pe: Pericarp. Pl: Plumule. Pr: Primary root.

A seed awakened from the dormancy begins to germinate within two days after the sowing, at 30–35°C, on the seed bed.

A seed (caryopsis) which is on the point of germinating swells and its volume increases by the absorption of water compared with that of a dry caryopsis.

Soon the growing embryo bursts its covering near the base of the caryopsis; the first portion of it to emerge is the dilated coleorhiza, which tears the pericarp and makes a longitudinal slit (Fig. 4, A).

Without delay the plumule exposes itself to the opposite direction (Fig. 4, C).

Later, a number of long unicellular hairs, resembling root-hairs in form and function, often arise from the cells of the coleorhiza.

After the coleorhiza has grown about 0.5 mm long, the enclosed primary root bores through it, usually on one side (Fig. 4, D). The primary root elongates much more quickly than the plumule (Fig. 4, C, D).

As the spikelet is protected with glumes it is not usual that in the early stages of germination the plumule and the coleorhiza can be seen perfectly. If the glumes are removed these become to visible.

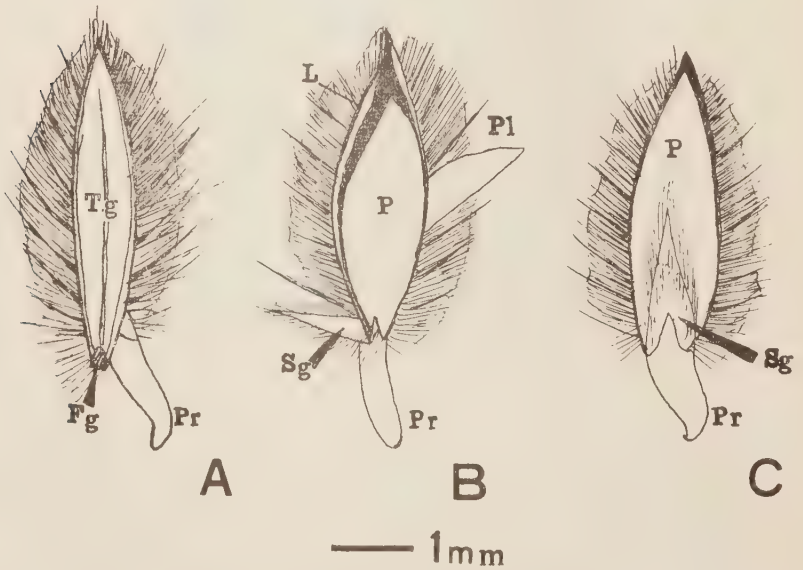

Fig. 5. Germination stage of a spikelet.

Fg: First glume. L: Lemma. P: Palea. Pl: Plumule.
Pr: Primary root. Sg: Second glume. Tg: Third glume.

The glowing plumule pulls apart the hull from the apex of the spikelet and appears on one side of the middle part of the spikelet (Figs. 5 and 6).

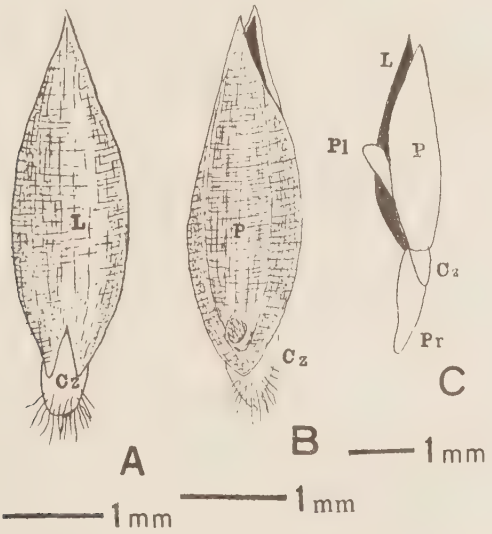

Fig. 6. Germination stage of a spikelet; glumes were removed.
Cz: Coleorhiza. L: Lemma. P: Palea. Pl: Plumule.
Pr: Primary root.

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Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11, No. 1
August 30, 1957

A second look at the ants of the *Camponotus herculeanus* group in Eastern Asia

Keizô Yasumatsu and William L. Brown, Jr.

Only six years ago, we reviewed the Palaearctic members of the *Camponotus herculeanus* group, with special reference to the forms of eastern Asia (Yasumatsu and Brown, 1951). The most important taxonomic decisions then reached concerned the groups *herculeanus-japonicus* and *obscuripes-hemichlaena*. We decided that *C. japonicus* G. Mayr was a temperate Asian race of the boreal *C. herculeanus* (Linnaeus), and that *hemichlaena* Yasumatsu et Brown was a south Japanese race of *C. obscuripes* G. Mayr, following the concepts of population systematics as summarized by E. Mayr (1942).

Within two years, however, the concept of subspecies as applied to animal systematics came into serious question (Wilson and Brown, 1953), so that a reappraisal of the *C. herculeanus* allies in eastern Asia was clearly called for. Such a reappraisal became possible under unusually favorable circumstances when, in 1956, Yasumatsu was able to visit the United States under the auspices of the Ministry of Education of the Japanese Government and had the opportunity to discuss the situation with Brown directly for the first time. At this meeting, we both had a chance to examine and consider additional information and material bearing on the *herculeanus*-complex problem, and as a result, we are able to offer a new interpretation that we believe to be more accurately representative of the natural relationships of the forms concerned.

A.—*C. herculeanus, japonicus* and *atrox*

In our 1951 paper, we offered evidence for the conspecificity of *japonicus* and *herculeanus*. This evidence consisted largely of the

demonstration that intergradient forms existed in a broad zone running through northern China, Manchuria and neighboring lands, as well as in the high mountains of Japan, Korea and China. Putative intergrades were in our material nothing like so abundant as "pure" *herculeanus* and *japonicus*, a fact that makes it now possible, at least for the sake of argument, again to consider the two "extreme" forms as separate species. However, if we are to make such a separation more than a point of rhetoric, the samples we formerly called intergrades must be explained.

The *japonicus* extreme differs from the *herculeanus* extreme as follows.

japonicus	*herculeanus* (E. Asia)
Color normally black overall, though head of major worker may be more or less dark red. Sculpture coarse, especially of gaster, dense, opaque. Median lobe of clypeus advanced distinctly beyond lateral lobes. Gastric pubescence long, coarse, abundant.	Color normally black, with more or less red on alitrunk and legs. Sculpture fine, gastric dorsum more shining. Median lobe of clypeus approximately even with lateral lobes. Gastric pubescence short, usually not surpassing the posterior segmental borders.

As already mentioned above and in our 1951 paper, the great majority of available eastern Asian samples can be determined unequivocally as either *japonicus* or *herculeanus* on the basis of the characters cited. There may also be a difference in head shape, but the analysis of this character would require the measurement of a great many specimens, including the largest sizes of workers, which we do not have in sufficient numbers from the *herculeanus* (northern) part of the range. In 1951, we felt that the "intergrades," found mostly at low altitudes, indicated a zone of secondary intergradation between two subspecies. We know now that the intergrade samples (workers and females) we have actually seen do not form a continuum, but instead seem to fall into three classes. These classes exclude characterizations from the literature, to which we unconsciously gave too much weight in 1951.

Class I.

North Chinese and Manchurian specimens, essentially *japonicus*, but often with the clypeal lobe slightly less noticeably projecting and with gastric pubescence a bit more dilute, as compared to Japanese examples. Larger workers sometimes have the head more or less reddish in color, in life as well as in dried Museum specimens. This is the form Emery described as var. *aterrima*; specimens in the MCZ from the southern Urals ("var. *saxatilis*" Ruzsky) agree well with north Chinese

"*aterrima*." Sculpture, pubescence and color are clearly very close to those of *japonicus*, and there is no convincing, direct evidence known to us to suggest that samples of this class occur at the same exact sites with, and there grade into, any more *herculeanus*-like class. Variation in projection of the clypeal lobe is a character most difficult to evaluate, since it is slight at best, and since it is allometrically controlled in the very size-variable worker-female castes of this complex. It seems that the character is not especially well correlated with density of pubescence or any other feature, and it is not even possible to be sure that specimens from Japan have a more strongly projecting lobe on the average than, say, samples from northwestern China.

It is our opinion now that Class I belongs to *C. japonicus*, and that it is in fact not far from typical for this species. The most conspicuous variate, gastric pubescence, seems to show slight reduction in peripheral areas (i.e., those away from Japan and central China), and not just in the direction of *herculeanus*-inhabited areas. In fact, samples of both species from Sikang Province (at different altitudes) are sharply distinct. The mountains of Sikang form the western scarp of the Tibetan Plateau, here rising sharply from the Red Basin of Szechuan. The *japonicus* samples from this general area of west-central China collected by D. C. Graham and by Brown show no tendency to vary toward *herculeanus* despite the proximity of the latter species here.

So far as can be determined from the many explicit records known to us, *C. japonicus*, including Class I samples, is always a soil nester in eastern Asia. The exact geographical range in full remains to be worked out. In addition to the Volga-Urals populations, there are presumably solid records such as that of Eidmann (1942), who noted *C. japonicus* from 2700 m. at Turbaling, N. W. Himalaya, taken by the Nanga Parbat Expedition. The Siberian records, mostly in Russian publications, are of little use to us, due to their generally obvious taxonomic and nomenclatorial confusion in dealing with this complex. Actually, we have little reliable and explicit information about the northward extent of the *C. japonicus* distribution, where it is sympatric with *C. herculeanus*, if anywhere, and how it behaves toward the latter if the two are in direct contact.

Class II.

Samples falling in this class are now considered by us to represent probable melanic examples of *C. herculeanus*. They agree with *herculeanus* in all characters except their color, which is nearly or completely black throughout. A sample from the Southern Japanese Alps contains some workers with the alitrunk entirely black, but others

with the extreme posterior part of the alitrunk red; these last examples are very similar to many Canadian and Alaskan samples in color as well as in other respects.

Apparently, the *sachalinensis* of Forel and some later authors is this melanic *herculeanus* form. Other melanic *herculeanus* populations are known from Italy and the western United States (see below). The black *herculeanus* samples can be distinguished from *japonicus* by sculpture, much less well developed pubescence, and by the short clypeal lobe. No intergradation to *japonicus* is known, despite intensive collecting in the mountains of Japan. It seems clear to us now that our category of the 1951 paper (p. 36): "*japonicus*...(b) Black intergrades to *herculeanus*" wrongly includes the present Classes I and II without distinction. Actually, these two classes seem to be examples of color convergence, not intergradation. Without seeing types, it is impossible to be sure whether varieties *jakutica*, *sachalinensis* and *manczshurica* belong to Class II, to mixtures of Classes I and II, or to something completely different from anything we have seen.

Class III.

This is equivalent to our 1951 *japonicus* category "(c) Intergrades to *herculeanus* with red alitrunk" (pp. 37–38). The prior name, which we shall apply here, is *atrox* Emery. The few specimens actually available to us are samples of a single nest series from Mt. Kongo in central Korea, from a single depauperate nest taken in Shansi Province by Yasumatsu, and a single major worker from "Eastern Tomb," presumably near Peking, in the MCZ Collection. These specimens have the sculpture and pilosity of *japonicus*, but the alitrunk is red, not black. The clypeus, for what this character is worth, seems to be intermediate between those of "typical" *japonicus* and *herculeanus*.

Yasumatsu found only the single small nest in Shansi, although he found a great many nests of *japonicus* during his collecting in this province. In neighboring Shensi Province, despite extensive collecting, Brown found only *japonicus* at Hanchung, Pao Cheng, Miao Tai Tze in the Tsinling Range, and on the Wei River Plain, and never saw *atrox*, either in this province or elsewhere in western and north-western China. Other published records referring to this form are from northern Korea and from Jehol. It seems reasonably clear that such a large ant, conspicuous by virtue of its color, must be rather uncommon and sporadic in China, Korea and Manchuria, at least, to have yielded so few and scattered records. That *atrox* is not known from the Japanese highlands, despite intensive collecting, is significant, especially when one considers that such experienced ant collectors as Yano, Teranishi, Yoshioka, Morishita, Silvestri, Okamoto and others have worked this area over a long period.

From our present knowledge, the true status of *atrox* is impossible to determine with any finality. Three possibilities seem worth considering.

(1) *Atrox* may be a northern or upland variant of *japonicus*. This has against it the apparent intermediate nature of the clypeus, if this character may be trusted. Also, *atrox* has not been found in Japan, although *japonicus* is of course abundant there. Where it occurs, *atrox* is apparently within the range of *japonicus*, so that the differences, if they are genetically controlled, would have to be under simple, all-or-none genetic control if applying to one and the same species.

(2) *Atrox* may be the result of occasional hybridization between *japonicus* and *herculeanus*. This possibility would be worth more consideration if the second putative parent, *herculeanus*, had ever been found unequivocally associated at even a single locality with *japonicus* and *atrox*. There appear to be no safe records of the three forms from any one locality. If hydridization occurs, one would except it to affect not only color, but also sculpture and pubescence, in the same areas where *atrox* was found; there exists no evidence to this effect. The lack of *atrox* in Japan does not mean too much, because there even the *herculeanus* are nearly or quite completely black.

(3) Possibly *atrox* is a species apart from both *japonicus* and *herculeanus*. If this is the case, *atrox* might represent the remnants of the parent stock (from which *japonicus* diverged), now caught between *japonicus*, expanding from the south, and *herculeanus*, occupying the far north. This third possibility may be the best one we can entertain for the time being, and so long as *atrox* seems to have a fair chance of being a good species, it will be best to recognize *Camponotus atrox* Emery (NEW STATUS) as a provisional full species.

The new examination of the *japonicus-herculeanus* "intergrades" shows that most of these forms can be placed to one or the other of the two species, and that they are probably not the result of interbreeding. At most, one could claim a limited amount of hybridization to account for some of these variants, but the availability of alternative explanations makes even this moderate hypothesis seem rather superfluous. At any rate, there is no good reason to continue to consider *japonicus* a "subspecies" of *herculeanus;* instead, we return to the original concept of Gustav Mayr, who described *Camponotus japonicus* as a good species. The certain synonyms of *C. japonicus* at this writing are: *aterrima* Emery, *saxatilis* Ruzsky, *sanguinea* Karawajew, *miltotus* Wheeler and *wui* Wheeler. Forms of uncertain affinities, but probably belonging to *C. herculeanus*, are: *jakutica* Karawajew, *sachalinensis* Forel, and *manczshurica* Ruzsky.

We urgently need to have a much clearer picture of the range and variation of *C. herculeanus* in central Asia and Siberia. The European *herculeanus* usually have all-red alitrunks, and when, as in the southern montane populations ("var. *nadigi*"), the alitrunk turns black, it does so in a rather even way, through increased melanization of the entire tagma. The American and Japanese samples, on the other hand, show more progressive melanization from front to rear; e.g., the propodeum alone may remain red, while the promesonotum is entirely black. Do both these tendencies exist in eastern Siberian populations, and if so, what happens where they merge or meet?

Another question surrounds the identity of the "*C. herculeanus eudokiae*" of Ruzsky, even the description of which remains unavailable to use. And then we have a nest series (MCZ) from Pskem, in the western Tien Shan, at an altitude of about 1000 m., with all-red alitrunk and other general features of *herculeanus*, but with the appressed gastric pubescence longer than usual, surpassing the posterior borders of the segments. Elsewhere, pubescence of similar, and even more extreme, development is found in the southern *herculeanus* populations of western North America ("race *modoc*"). We hope these problems have attracted or will attract the attention of Russian ant specialists.

B. *C. obscuripes* and *hemichlaena*

We described *hemichlaena* in 1951 as a provisional southern race of *C. obscuripes*, but additional information now available leads us to separate these two as good species, so that the former now is considered as *Camponotus hemichlaena* Yasumatsu et Brown (NEW STATUS). *C. hemichlaena* has a characteristic black prothorax, contrasting sharply with reddish color of the rest of the alitrunk. The species is widely distributed in Kyushu, Shikoku and southern Honshu. Many collections and observations made in these areas now agree in showing that *obscuripes* and *hemichlaena* frequently occur in the same or closely adjacent localities, but without producing intergrades.

We had already reported in 1951 that *obscuripes* occurs rarely at high altitudes in Kyushu. Now we find that both species occur in the same districts of eastern Shikoku. Generally speaking, *obscuripes* tends to occupy the higher altitudes, but altitudinal separation is not complete. Samples from Honshu indicate that the situation is much the same there in the southern part of the island; no intermediates are yet known from any of these areas. All colony series so far seen are "pure," either *obscuripes* or *hemichlaena*, never both. There are some indications of direct struggle between the two in eastern Shikoku, but their bionomics and competitive status have not yet been studied in any detail.

The circumstances suggest that *hemichlaena* is a southern derivative from the *obscuripes* stock that is now slowly replacing the latter from south to north and from lower to higher altitudes. The possibility is even more interesting when one considers that *C. japonicus* may be advancing northward against *C. herculeanus* in Asia, while the same may be true of *C. pennsylvanicus* De Geer vs. *C. herculeanus* in the eastern part of North America (Brown and Wilson, unpublished data). Conclusive demonstration of such northward expansion of warm-zone against cold-zone species would only be following the principles laid down in the zoogeographic writings of P. J. Darlington (1948 and in press).

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Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11, No. 1
August 30, 1957

Studies on the multiplication of OP_1 phage (*Xanthomonas oryzae* bacteriophage). 2

Interference phenomena in the multiplication between OP_1 and OP_{1t}, the growth temperature mutant obtained from OP_1*

Satoshi Wakimoto and Hazime Yoshii

Introduction

The multiplication of bacteriophage starts by the injection of the contents—desoxyribonucleic acid—of the phage adsorbed on the surface of the host cell. The penetrated DNA from the parent phage plays in the cell the principal role of the reproduction of new phages as the genetic marker.

When a bacterial cell is infected by two kinds of the virulent phages, the interference phenomena between the phages will take place. Phages are classified as the related with each other or as the unrelated on the standpoint of their serological reaction and morphological relationship. And the interference phenomena between two phages are different in accordance to their mutual relationships.

In the case of the unrelated phages, as it is reported with *Escherichia coli* phage α (T1), δ (T7) and γ (T2), the mutual exclusion phenomena and depressor effect have taken place.[5,6] Namely, in the case of simultaneous infection of two unrelated phages, the contents of one of the two phages penetrate and multiply, while the other phage cannot self-reproduce due to the exclusion of the previously penetrated phage. The excluded phage which adsorbed on the cell surface, in its turn, interferes or depresses the multiplication of the intruded phage.

In the case of the related phages, both two phages penetrate and multiply without any interference between them, if they simultaneously attack the same host cell. In the progenies, besides the reproduction

* Contribution from the Laboratory of Plant Pathology.

of normal phages of the characters of the respective parents, are produced the recombinants having the characters of both two parents as the results of recombination of the parent characters. When infection is unsimultaneous, lapse of time necessary for the excluding action of the prior phage against the succeeding phage is determined by the nature of the former. Interference phenomena of related phages were detailed in the experiments with T-even phages by Dulbecco.[8, 9]

The authors isolated a mutant —OP_{1t} phage—which differs in its optimum growth temperature from the wild type OP_1 (*Xanthomonas oryzae* phage). Many kinds of phage mutant such as host range mutant,[7, 13] plaque morphology mutant,[7, 12] adsorption cofactor mutant,[3] proflavine resistant mutant,[11] and heat resistant mutant[2] were reported. The OP_{1t} phage is a new type mutant and is named the growth temperature mutant.

The writers report the experiments on the interference of OP_{1t} to OP_1 phage multiplication studied by the single-burst experiment[4, 10] and the one-step growth experiment.[10]

Materials

OP_1 phage: A host specific bateriophage of *Xanthomonas oryzae*, the causal organism of the bacterial leaf blight of rice, and some physiological and biological characters of the phage were reported.[15]

OP_{1t} phage: The growth temperature mutant obtained from OP_1, and has the same serological and several other characters except in its optimum growth temperature with OP_1.

X. oryzae no. 60: Newly isolated strain obtained from *X. oryzae* no. 49.

CaVfCh medium: Vitamin free caseinhydrolysate medium with $CaCl_2$. The protocol of this medium was reported in the previous paper.[15] This medium is profitable for both OP_1 and OP_{1t} phage multiplications but is not so preferable for the multiplication of *X. oryzae* as the semiartificial medium.

Anti-OP_1 phage serum: The method to make this was also previously reported.[15] It reacts with both OP_1 and OP_{1t} phages in equal activity, and inactivates them with the same inactivation constant.

Method

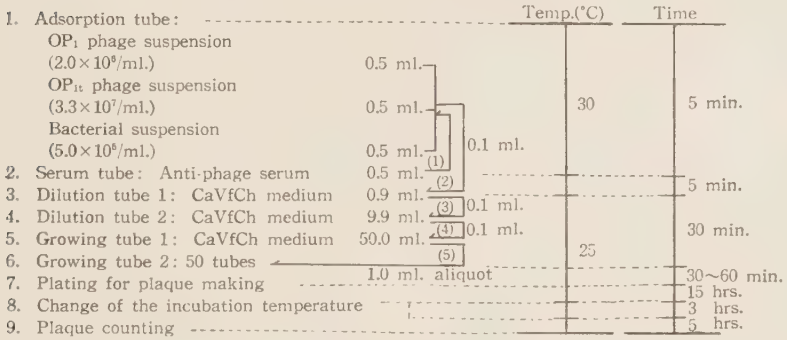
The single-burst experiment is the method with which the reproduced phages from each infected single bacterial cell are assayed quantitatively. If two kinds of phages attack a bacterial cell simultaneously or unsimultaneously, the interference, if present, between both phages in the same host cell will be cleared by this method. The

principal of this method is to divide the phage infected bacterial suspension to the test tube—growing tube 2 in Protocol 1—, before bacterial lysis takes place, so that each tube will contain, on the average, less than one cell of the infected bacteria. After the burst of all infected bacteria in each tube under definite environmental condition, the produced phage progenies are quantitatively assayed by usual plaque count method.

The procedures of the single-burst experiment used in this experiment are indicated in Protocol 1 and 2.

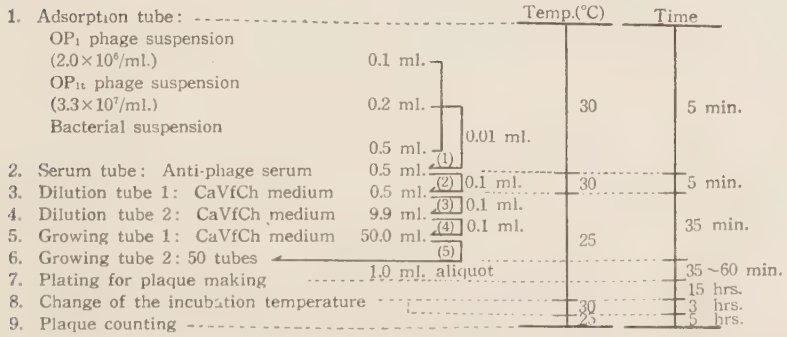
Protocol 1

Protocol of single-burst experiment (1)



Protocol 2

Protocol of single-burst experiment (2)

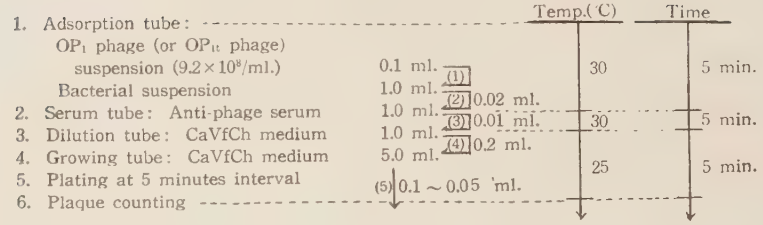


The method used for the one-step growth experiment of OP_1 and OP_{1t} phages is as Protocol 3.

The discriminating method of OP_1 from OP_{1t} phage by plaque morphology is as follows.

OP_1 phage forms clear plaques at 25°C. or 30°C. on *X. oryzae* no. 49 (Plate , figs. 1 et 3), while it forms indistinct plaques at 35°C. (Plate

Protocol 3
Protocol of one-step growth experiment



1, fig. 5) Namely, *X. oryzae* no. 49 cells infected with OP_1 phage are lysed prefectly under these temperatures unless at 35°C. While OP_1 phage forms clear plaques at 25°C., and cannot at 30°C. The ability of these phages to lyse the infected bacteria might be depressed by some unknown reasons under such high temperatures.

After plating the mixture of OP_1 and OP_{1t} phages by usual method, and placing the culture plate successively at 25°C. (15 hrs.), 30°C. (3 hrs.) and 25°C. (several hrs.), clear rings will appear in every plaque of OP_{1t} phages on account of lysis inhibition of the infected bacteria under 30°C., while there are normal plaques in the case of OP_1 phages. By the presence or absence of the rings in plaques, OP_1 and OP_{1t} phages are distinguishable from each other easily (Plate , figs. 2 et 4).

If the plate of the bacteria infected alone with OP_1 phages is incubated successively at 25°C., 35°C. and 25°C. each under the appropriate period the plaques having clear rings will appear as when OP_{1t} infected bacteria is incubated at 25°C., 30°C. and 25°C. successively. These are based on the fact that both phages differ from each other only phenomena in their optimum growth temperatures.

Results

The result obtained by conforming to Protocol 1 is showed on the table of Result 1.

Result 1
Result of single-burst experiment

Tube no.	No. of plaques		Tube no.	No. of plaques	
	OP_1	OP_{1t}		OP_1	OP_{1t}
1	0	0	6	0	22
2	15	9	7	2	32
3	20	14	8	4	24
4	6	9	9	2	34
5	13	19	10	3	27

11	7	26	31	0	44
12	0	39	32	0	27
13	0	1	33	0	43
14	0	0	34	0	0
15	26	26	35	0	43
16	0	2	36	10	15
17	13	3	37	17	2
18	0	0	38	0	16
19	17	16	39	43	23
20	0	2	40	1	20
21	0	20	41	7	23
22	0	5	42	1	28
23	7	3	43	9	37
24	0	0	44	11	13
25	14	17	45	0	32
26	6	7	46	0	53
27	0	40	47	6	23
28	0	31	48	47	26
29	0	35	49	0	39
30	19	13	50	0	4

Total tubes	Tubes containing bacteria infected with OP_1	Nos. of phages in tubes		
		OP_1	OP_{1t}	Total
50	26	326	489	815

Check (no. of free phages) per 0.1 ml. of dilution tube 2.

OP_1	OP_{1t}
2	6

The result obtained by conforming to Protocol 2 is showed on the table of Result 2.

Result 2

Result of single-burst experiment

Tube no.	No. of plaques		Tube no.	No. of plaques	
	OP_1	OP_{1t}		OP_1	OP_{1t}
1	0	57	6	0	81
2	0	85	7	0	81
3	3	59	8	0	88
4	0	118	9	0	112
5	0	70	10	7	120

11	1	95	31	4	69
12	22	54	32	0	100
13	0	90	33	0	100
14	0	56	34	7	120
15	0	90	35	0	140
16	21	75	36	37	112
17	0	96	37	28	90
18	0	92	38	0	95
19	0	120	39	1	140
20	0	58	40	0	150
21	0	110	41	0	95
22	15	140	42	14	65
23	15	96	43	0	120
24	7	82	44	16	70
25	25	45	45	0	120
26	0	79	46	0	70
27	0	112	47	0	70
28	13	160	48	15	200
29	29	102	49	1	170
30	0	67	50	0	190

Total tubes	Tubes containing bacteria infected with OP_1	Nos. of phages in 20 tubes		
		OP_1	OP_{1t}	Total
50	20	281	2064	2345

Check (no. of free phages) per 0.1 ml. of dilution tube 2.

OP_1	OP_{1t}
1	26

The multiplication formulae of OP_1 and OP_{1t} phages on *X. oryzae* no. 60 resulting from one-step growth experiment conforming Protocol 3 are given in Result 3.

Result 3

Result of one-step growth experiment under 25°C.

	Latent period (min.)	Rise period (min.)	Average burst size
OP_1	45	20	25 (22–28)
OP_{1t}	40	20	25 (22–28)

Discussion

As stated in the introduction, the interference phenomena between two kinds of phages on the site of reproduction are different according to whether the phages are related or not. When serologically or morphologically related phages attack simultaneously the same host cell, both phages will penetrate and multiply accompanied with some recombinations.

OP_{1t} is the phage derived from the wild type OP_1 and is related to the latter with the exception of its optimum growth temperature. It will be expected therefore that simultaneous penetration and multiplication of both two phages in the same host cell will occur when they are inoculated simultaneously. Protocol 1 is the single-burst experiment carried out with the expectation to confirm the existence of above phenomena and to confirm the possibility of alternation of phage reproducing ability of the double infected bacteria with that of the simple infected bacteria.

The distribution pattern of infected bacteria to 50 growing tubes in Result 1 is discussed with Poisson's distribution.

$$P(r)=\frac{n^r \cdot e^{-n}}{r!}$$

$P(r)$: Proportion of tubes containing bacterial cells infected with phages in number (r).

n: Average number of infected bacteria per tube.

The following results were obtained with OP_1 phage.

	No. of tubes (50·P)	No. of bacteria infected with OP_1 (r · 50 · P)
P (0) 0.48	24	0
P (1) 0.353	18	18
P (2) 0.131	7	13
P (3) 0.032	2	5
P (4) 0.006	0	0
		Total 36

where $n=0.74$ because $P(0)=\frac{24}{50}=e^{-n}=e^{-0.74}$

Namely, out of 26 tubes which produced OP_1 phage, 18 tubes had contained one infected bacterial cell each, 7 tubes 2, 2 tubes 3, and no tubes were found to contain 4 or more bacterial cells infected with the phage. So, the total number of bacteria infected with OP_1 phage in 26 tubes was calculated at 36. The average burst size of OP_1 phage

in this experiment were therefore obtained by dividing 326—total yields of OP_1 phages by 36 —the total infected bacterial number of 26 tubes.

$$N_{op_1} = \frac{326}{36} = 9$$

As to OP_{1t} phage, by dividing total yields of 489 in 26 tubes in Result 1 by total OP_1 infected bacteria of 36, the average burst size was obtained.

$$N_{op_{1t}} = \frac{489}{36} = 13.6$$

Considering it as a whole, 26 tubes containing 36 infected bacteria which produced both OP_1 and OP_{1t} phages; the total phages produced amounted to 815. So the average burst size of the mixed phages (N) was calculated as follows:

$$N = \frac{815}{36} = 22.6$$

These figures are nearly equal to those which were obtained from Protocol 3, Result 3. In other words, there are scarcely any differences on the phage producing ability of the bacteria when the case of mixed infection in compared with that of simple infection.

Protocol 2 is the experiment in which the proportion of the number of OP_{1t} phages to that of OP_1 is changed. From the table of Result 2, the average burst size of the bacteria regarding OP_1 phage is about 9 which is the same as that in the case of Result 1 obtained from the experiment Protocol 1. In other words, in the case of mixed infection, so far as this experiment in concerned, the rate of phage producing ability of the bacteria is not affected even if the ratio of the particle number of the inoculated phages is changed.

Finally, comparing the refults in the tables of Result 1 and 2 with those of Result 3, it is reasonable to conclude that the partition in constant proportion to the reproducing ability of the host bacteria of OP_1 and OP_{1t} phages, in the case of mixed infection, is due to the difference in the latent periods of both phages.

Summary

OP_{1t} phage, one step mutant of OP_1 (*Xanthomonas oryzae* phage), is found to be a new type mutant. It is called a growth temperature mutant because its optimum growth temperature (25°C.) differs from that of the wild type (30°C.).

When these two phages attack simultaneously the same host baterial cell, they both penetrate and multiply.

	Latent period (min.)	Rise period (min.)	Average burst size
OP_1	45	20	25 (22–28)
OP_{1t}	40	20	25 (22–28)

The phage producing potency of the cell is divided not in accordance with the different multiplicity of the inoculated phages but with their faculties or reproducing speed and the cell released both types of phage progenies in a definite proportion.

Scarcely any difference on the average burst size between the case of mixed infection and that of simple infection is found.

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Explanation of Plate 1

Fig. 1. Plaques formed by OP_1 phages, incubated for 15 hrs. at 30°C.

Fig. 2. Plaques formed by OP_{1t} phages, incubated for 15 hrs. at 25°C., 3 hrs. at 30°C. and 3 hrs. at 25°C. successively.

Fig. 3. Plaques formed by OP_1 phages, incubated for 15 hrs. at 25°C., 3 hrs. at 30°C. and 3 hrs. at 25°C. successively.

Fig. 4. Plaques formed by OP_1 mixed with OP_{1t} phages, incubated for 15 hrs. at 25°C., 3 hrs. at 30°C. and 3 hrs. at 25°C. successively. These plaques with rings on the upper left side are formed by OP_{1t}. While the others are formed by OP_1.

Fig. 5. Plaques formed by OP_{1t} phages, incubated for 15 hrs. at 30°C.

Fig. 6. Plaques formed by OP_{1t} phages, incubated for 10 hrs. at 30°C. and 5 hrs. at 25°C. successively.

Studies on the multiplication of OP_1 phage

Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11, No. 1
August 30, 1957

On the characteristics of the transparent fluid

II An electrophoretic study of proteins of the transparent fluid

Hisayoshi Nishiyama

The transparent fluid of the cock is the fluid which flows from its accessory reproductive organs and is added to the vas deferens semen simultaneously with the ejection of it. The fluid, then, corresponds to the secretion of the accessory reproductive organs in mammals (Nishiyama, 1955). However, in the previous paper the writer (1954) reported that the origin of the transparent fluid, unlike the secretion of accessory reproductive organs in mammals, lay in theblood.

It was of interest to investigate whether or not the protein constituents of the transparent fluid were similar to those of the blood serum of the cock. For this paper, the composition of transparent fluid was analyzed by electrophoresis and was compared with that of the blood serum of the same bird.

This investigation was carried out under the direction of Professor Masaharu Tange.

Materials and method

In the cock, the vas deferens semen is ejected from the openings of vasa deferentia and the transparent fluid flows out simultaneously with the ejection of vas deferens semen from the lymphfolds of both sides. Then, in order to collect the transparent fluid, a cock which had been obstructed from contamination of the semen in vas deferens, by means of binding the anterior vasa deferentia with surgical threads and destruction of the openings of vasa deferentia with electric cautery was used as in the previous paper (Nishiyama, 1954). The transparent fluid was collected by means of abdominal massage method (Burrows

and Quinn, 1937) from a cock mentioned above. In collection, the outside of the anus was cleansed and a mass of cotton was inserted into the rectum in order to block any contamination. Soon after the collection, dilute gelatinous substance or substances considered to be a matter similar to fibrin appeared in the collected transparent fluid, as reported in the previous paper (Nishiyama, 1955). This fibrin was removed and the fluid without fibrin was used. The amount of the transparent fluid which was able to be collected by abdominal massage was 1.2 ml. on the average. The collected fluid was pooled in a test tube at 0°C until the total amount reached 7—10 ml. As the protein concentration of the fluid was only 0.4 per cent, the fluid could not be analyzed in this state and it needed to be condensed. Then, the pooled fluid samples were condensed to 3 ml. by dialysis in 30 per cent arabic gum solution which was placed in a refrigerator at 4–6°C. It was more suitable to make the 30 per cent arabic gum solution with veronal buffer (0.05 M Sodium diethylbarbiturate, 0.01 M diethylbarbituric acid) rather than make it with distilled water. When the solution was made with water, some of the precipitation arose in the course of condensing although this precipitation was dissolved away by dialysis with a veronal buffer which was done before analysis; on the other hand, when a veronal buffer was used as solvent, none of the precipitation arose in the course of condensing. The protein concentration rose 1—1.4 per cent in this manner. The concentrated fluid was placed in cellophane tubing and dialyzed against the veronal buffer mentioned above (pH 8.6, ionic strength 0.06) at 4—8°C for a period of 1 or 2 days. Electrophoretic analysis was also performed with veronal buffer, employing the Hitachi Electrophoresis Apparatus with micro cell. Electrophoresis was carried out at five milliamperes and approximately 140 volts for about 3600 seconds. The relative mobilities of various components, which are the relative percentage of displacements of each peak compared with the displacement of component 1 as 100 per cent, were calculated from an electrophoretic diagram, and from these mobilities corresponding components of each diagram were identified. The relative per cent composition of the components was determined by the method of Longsworth (1942).

Blood samples were obtained from the brachial vein of the same bird from which was collected the transparent fluid samples. Just after collection, the blood sample was placed in an incubater at 38°C and when clotting appeared to be complete, the sample was placed into a refrigerator and allowed to stand for about 16 hours to separate the blood serum. The serum was dialyzed with a veronal buffer and analyzed electrophoretically.

The protein concentration of both serum and the transparent fluid was determined with the Hitachi protein refractometer and the fluid

protein concentration was again ascertained by the semi-micro Kjeldahl method. The concentration of the fluid was determined as 0.38 and 0.43 per cent on the average with the refractometer and Kjeldahl methods. On the other hand, serum had an average concentration of 5.1 per cent. The final protein concentrations of serum and the fluid were 2.0—3.0 per cent (on an average, 2.5 per cent) and 0.4—0.8 per cent (on an average, 0.6 per cent) respectively.

Results and discussion

The blood protein components of the fowl have been studied by Tiselius electrophoretic method or by the method of filterpaper electrophoresis (Sanders et al., 1944; Deutsh and Goodloe, 1945; Moore, 1945, 1948; Brandt et al., 1951; Clegg et al., 1951, 1953; Common et al., 1953; McKinley et al., 1953). In general, it has been noticed that the plasma of the fowl contains 6 components and serum contains 5 owing to the lack of fibrinogen. These fractions are designated usually as albumin, α_1 globulin, α_2 globulin, β globulin, fibrinogen and γ globulin, by analogy with human plasma fractions.

In the preliminary experiments, the writer photographed many plasma and serum patterns of the white Leghorn cocks which were analyzed in the boric acid buffer after Brandt et al. (1951), and plasma patterns were identical to those represented by Sanders et al. (Fig. 1 a).

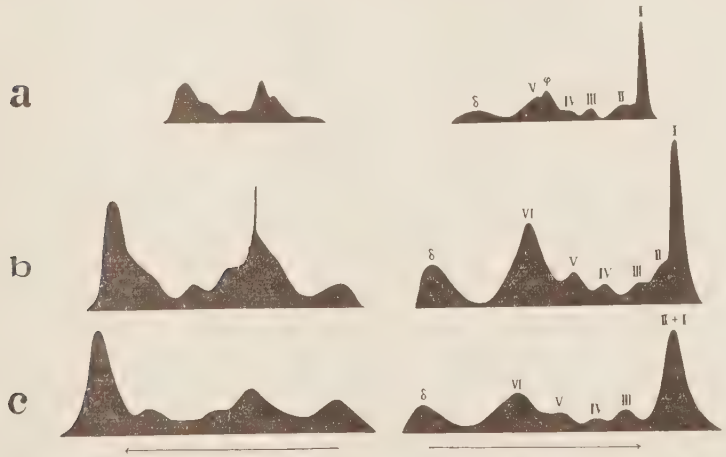

Fig. 1. Electrophoretic patterns of plasma, serum and transparent fluid of the cock. a. Plasma pattern analyzed with boric acid buffer. b. Serum pattern analyzed with veronal buffer. c. Pattern of transparent fluid analyzed with veronal buffer.

In the cock serum, it lacked a fibrinogen peak and there were 5 peaks. Component 1 to 5 in order of decreasing mobility was designated respectively as albumin, α_1, α_2, β and γ globulin, from the result of this experiment. On the other hand, in the patterns of the cock serum which were analyzed with veronal buffer as in this experiment, 6 peaks were observed in almost all cases, including a new peak situated between component one and two, in respect of their relative mobilities (Fig. 1 b, Table 1). McKinley et al. (1953) who studied the chicken serum by paper electrophoresis, demonstrated 6 components and they designated these fractions as albumin, α_1, α_2, α_3, β and γ globulin. Then, the fractions which had been considered as component 2 (α_1) and 3 (α_2) in the patterns which were analyzed with boric acid buffer it might be more suitable to designate as component 2 plus 3 ($\alpha_1+\alpha_2$) and 4 (α_3) respectively.

Table 1. Electrophoretic analyses of transparent fluid and blood serum in the cock.

	Relative per cent composition						Relative mobilities (%)					
	1	2	3	4	5	6	1	2	3	4	5	6
Transparent fluid	52.4	–	5.8	2.9	6.0	32.9	100	–	82	71	56	40
	50.4	–	8 6	3.3	6.3	31.4	100	–	82	69	60	37
	46.8	–	9.8	5.6	8.4	29.4	100	–	81	69	57	38
Blood serum	31.0	8.6	4.8	6.0	9.8	39.8	100	93	81	70	57	40
	33.4	7.3	5.0	5.9	8.9	39.5	100	90	83	71	58	42
	30.1	10.2	6.4	6.0	10.5	36.8	100	91	81	70	58	39
	27.0	8.6	6.9	5.6	11.1	40.8	100	93	83	69	57	38

The protein concentration of the transparent fluid was only 0.4 per cent as mentioned before and it was much lower than that of blood serum. However, this fact does not deny the assumption that the transparent fluid is a fluid similar to lymph. Because the protein content of lymph is less than blood serum and the concentration varies markedly in different regions of the body. Lymph coming from the liver has a relatively high protein content, about 5 per cent, lymph obtained from subcutaneous tissues, however, contains less than 1 per cent protein under normal conditions (Cantarow and Schepartz, 1954). Low protein concentration of the transparent fluid is presumably based on the facts that the fluid was generated from the lymphoid tissue or the vascular body which laid subcutaneous and that the amount of the fluid generated was very large in a short time (Cf. Nishiyama, 1955).

The relative per cent composition of the transparent fluid compared with blood serum is presented in Table 1 and Fig. 2, and the average value for each component is as follows: component 1 and 2, 50 per cent; component 3, 8 per cent; component 4, 4 per cent; component 5, 7 per cent and component 6, 31 per cent. Assuming the value of component 2 of the transparent fluid was the same as that of blood serum, i.e., 9 per cent, the relative per cent composition of the component 1 (albumin fraction) became 41 per cent. Still, the largest difference between fluid and serum was seen in component 1.

It is said that the relative portion of the protein of the lymph is almost the same as that of blood serum, although the albumin fraction of the lymph is somewhat larger in amount than blood serum (Hirai, 1953).

These evidences mentioned above support the assumption that the transparent fluid is a fluid similar to lymph.

In the previous paper (Nishiyama, 1955), the writer had presumed that the very small amount of secretion from the epithelial cells of the lymph-folds might be added to the lymph to make up the transparent fluid. The result of this experiment, however, revealed that the secretion to be added, if any, was negligible.

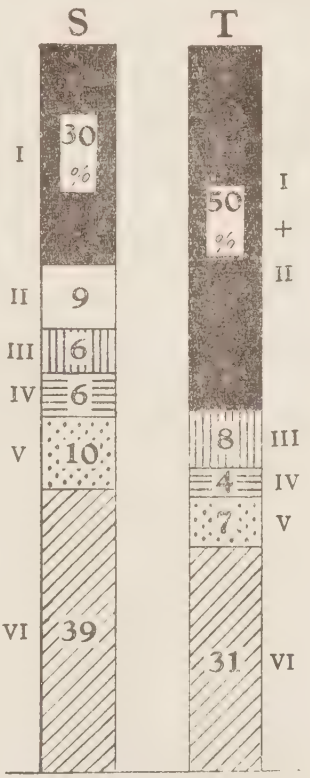

Fig. 2. Average value for each component of serum and transparent fluid.
S=Serum. T=Transparent fluid.

Summary

1. Transparent fluid, accessory reproductive fluid of cocks, contained all protein fractions which were contained in blood serum. The protein concentration of the fluid was, however, very low, i.e., 0.4 per cent on an average.

2. The relative per cent composition of the protein fractions of

the transparent fluid was similar to that of blood serum, although the albumin fraction of the fluid was larger than serum.

3. The results of this experiment support the assumption that the transparent fluid is a fluid similar to lymph.

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Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11 No. 1
August 30, 1957

A new species of a goby with a synopsis of the species of the Genus *Luciogobius* Gill and its two allied genera[1,2]

Yosie Dôtu

The author collected over one hundred specimens of a gobioid fish on the coasts of the Amakusa Islands, Kumamoto Prefecture, and the Tusima Islands, Nagasaki Pref. both in Kyūshū, which seemed to belong to the Genus *Luciogobius* Gill, and to be new to Science.

In this paper is given an account of the new species above mentioned with a synopsis of seven species of gobies belonging to the Genus *Luciogobius* Gill and its allied genera, *Inu* Snyder and *Expedio* Snyder.

Before going further, the author wishes to express his gratitude to Prof. Keitaro Uchida for his kind guidance and advice in this research and the preparation of this paper. Acknowledgements are also due to Dr. Ichiro Tomiyama and Dr. Tokiharu Abe who kindly offered facilities for consulting important literature and for the reexamination of the related specimens preserved in the Faculty of Science of Tokyo University: to Prof. Kiyomatsu Matsubara for facilities for consulting important literature: to Mr. Hiromu Ohashi and Mr. Satoshi Mito for their kind assistance in collecting specimens: and to Mr. Yoichi Syojima in the preparation of photographs.

Genus *Inu* Snyder

Inu koma Snyder
Snyder, J. O. 1909; Koumans, F. P. 1931; Matsubara, K. 1955.

[1] Contribution from Fisheries Laboratory, Faculty of Agriculture, Kyūshū University.

[2] A part of this research is owing to the Aid for Fundamental Scientific Research of the Ministary of Education. (Keitaro Uchida)

Genus *Luciogobius* Gill

Luciogobius guttatus Gill
Gill, T. 1859; Günther, A. 1861; Jordan, D. S. and Snyder, J. O. 1901; Regan, C. T. 1905; Koumans, F. P. 1931; Tomiyama, I. 1936; Regan, C. T. 1940; Matsubara, K. 1955.

Genus *Expedio* Snyder

Expedio parvulus Snyder
Snyder, J. O. 1909; Matsubara, K. 1955.

Key to the seven species belonging to three allied genera

A. Ventral fin present
  B. Posterior part of body scarly—Genus *Inu* Snyder
    C. No. dermal ridge on head .....*Inu ama* Snyder
    CC. Dermal ridge on head .....*Inu koma* Snyder
  BB. Scale absent—Genns *Luciogobius* Gill
    D. Ventral fin moderate
      E. Eye normally developed, numerous dark spots on body
        F. Fleshy barbels under eye.....*Luciogobius saikaiensis* n. sp.
        FF. No. fleshy barbel under eye.....*Luciogobius guttatus* Gill
      EE. Eye reduced, not or slightly pigmented
        .....*Luciogobius albus* Regan
    DD. Ventral fin a small flap.....*Luciogobius elongatus* Regan
AA. Ventral fin absent—Genus *Expedio* Snyder .....
  .....*Expedio parvulus* Snyder

Table 1. Measurements of the seven species of the three genera.

Specific name	Head in length	Depth in length	Dorsal fin rays	Anal fin rays	Pectoral fin rays	Vertebrae*
Inu ama Snyder	3.3	5.6	9	10	–	–
Inu koma Snyder	3.4～4.0	6.0～7.5	11～12	10～11	17～20	30～31
Luciogobius saikaiensis n. sp.	3.5～4.0	8.0～9.5	8～10	9～10	18	32
L. guttatys Gill	4.5～6.0	6.5～10.5	11～15	8～10	17～18	35～38
L. albus Regan	3 5～5.5	8.0～13.0	10～11	10～12	12～14	31～34
L. elongatus Regan	7.0～8.0	10.0～12.0	7～9	8～10	12	42
Expedio parvulus Snyder	5.5～6.5	11.0～15.0	9～10	11～12	12～13	42～44

* counted on the X-ray photographs.

Inu ama Snyder

(Japanese name, Ama-haze)

Snyder, J. O. 1909; Matsubara, K. 1955.

Synonym—*Luciogobius guttatus ama* (Snyder), Tomiyama, I. 1936.
Locality—Misaki, Kanagawa Pref. (Snyder, J. O. 1909); Sirahama, Tiba Pref. (Sakamoto, K. 1932).

No. specimen was examined in this study.

Inu koma Snyder

(Japanese name, Koma-haze)

Snyder, J. O. 1909; Matsubara, K. 1955.
Synonym—*Luciogobius guttatus koma* (Snyder), Tomiyama, I. 1936.
Locality—Misaki, Kanagawa Pref.; Oosima, Tokyo Capt.; Simoda, Sizuoka Pref. (Tomiyama, I. 1936); Kominato, Tiba Pref. (The author's collection); Fusan, Korea (Mori, T. 1952).

Eighteen specimens, 12~40 mm in total length, were examined in this study. Some of them had been studied by I. Tomiyama (1936). The life history and bionomics will be reported in another paper.

Luciogobius saikaiensis n. sp. (Fig. 1 and Plate 2)

(Japanese name, Hige-mimizu-haze)

Head 3.5 in standard length, depth 8.3, depth of caudal peduncle 11, length of caudal peduncle 5; eye 8 in head, snout 4, interorbital space 5.4, width of head 1.4; width of body 1.2 in depth; dorsal 10; anal 10; pectoral 18. Vertebrae 32.

Fig. 1. *Luciogobius saikaiensis* n. sp.
Male adult, 35 mm in total length, one of the paratype specimens.

Body cylindrical anteriorly and moderately compressed posteriorly. Head broader than the body and considerably depressed, and muscles of side and top of head greatly bulging, thus forming a deep trough behind eye on the occiput. Interorbital space broad, slightly concave with narrow transverse fleshy ridge. Maxillary extending backward to a point beneath posterior part of eye. Mouth-cleft almost vertical anteriorly, but horizontal posteriorly. Teeth very small, arranged in

narrow bands on both jaws, outmost ones enlarged. Tongue broad, deeply notched and free anteriorly. Gill-cleft restricted to the side. Five fleshy barbels below eye, and a large one in front of eye. A pair of barbels at the snout. Nasal canal tubular, posterior one near eye. A ridge with a row of large sensory organs across the snout and along the side of head backward to below eye. A similar organ on the lower jaw, cheek, and occiput. Head and body wholly nacked. Spinous dorsal absent. A slight median depression on the back before the dorsal with a row of minute plicae on its sides, those plicae being preceded by a slight, median ridge. Dorsal and anal opposite, inserted midway between bases of ventral and caudal; base of dorsal short and subequal to anal, both lengths about 2 in head. Dorsal somewhat higher than anal, both fins never reaching to the base of caudal when depressed. Membranes of dorsal and anal fleshy. Caudal rounded, 1.5 in head. Pectoral rounded, slightly pigmented, with 18 rays, the uppermost and lowest rays free and serrated. Ventral I, 5; anterior part thickened, posterior part with weak rays and fleshy membrane, and free posteriorly. A small genital papilla closely behind anus.

Color in spirit blackish, ventrally and caudad with white spots, vertical fins whitish with dark stripes across fin rays.

Holotypes—41 mm in total length, 35 mm in standard length male adult; from the sea-shore of Tomioka, the Amakusa Islands, Kumamoto Pref., Kyūshū.

Paratypes—Over 140 specimens, 5~41 mm in total length; from the sea-shores of Tomioka and Tuzizima, both in the Amakusa Islands, and Sasuna and Hidakatu, both in the Tusima Islands, Nagasaki Pref.

The type specimens are deposited in the Fisheries Laboratory, Faculty of Agriculture, Kyūshū University.

The life history and bionomics of this species will be reported in another paper.

(*Saikaiensis* means from West Sea District of Japan.)

Luciogobius guttatus Gill

(Japanese name, Mimizu-haze)

Gill, T. 1859; Günther, A. 1861; Jordan, D. S. and Snyder, J. O. 1901; Tomiyama, I. 1936; Regan, C. T. 1940; Matsubara, K. 1955.

Synonym—*Luciogobius guttatus guttatus* Gill (part), Tomiyama, I. 1936.

Locality—Japan throughout, Korea, and North China.

Over one thousand specimen, 7~72 mm in total length, from sea-shores of various districts of Japan, were examined in this study.

The life history and bionomics were reported in another paper (Dôtu, Y. 1957).

Luciogobius albus Regan[3]

(Japanese name, Dôkutu-mimizu-haze)

Regan, C. T. 1940; Matsubara, K. 1955.

Synonym—*Luciogobius guttatus guttatus* Gill (part), Tomiyama, I. 1936.
Luciogobius pallidus Regan, Regan, C. T. 1940.

Locality—Daikoniza, Shimane Pref., from a subterranean cave and wells; Gobô, Wakayama Pref., from wells; Uwazima City, Ehime Pref., from a well; Misaki, Kôti Pref. from a well (Tomiyama, I. 1936).

Head 4 in length; depth 9; depth of caudal peduncle 12; D. 10; A. 10; P. 13, V. I, 5; Vertebrae 32.

Body cylindrical anteriorly, caudal peduncle moderately compressed. Head depressed. Eyes vestigial beneath skin. Tactile organs of head not so highly developed as the North American Blind Goby, *Typhlogobius californensis* Steindachner (Hubbs, C. L. 1927) and *Lethops connectens* Hubbs (Hubbs, C. L. 1920). Mouth large, teeth minute, in narrow bands on both jaws. Head and body nacked, anterior nostoril with a short tube. No barbel. All fins large. Anal fin inserted directly below the second ray of the dorsal. Dorsal fin higher than depth. Anterior fraenum of ventral making a deep pocket. No free ray in pectoral fin. Color in spirit yellowish white, unpigmented.

Holotype—43 mm in total length, 37 mm in standard length, from Daïkonzima. Preserved in the Faculty of Science, Tokyo University. Specimen No. 25,693.

Paratypes—Thirteen specimens, 27～87 mm in total length, from above mentioned various localities. All the specimens are deposited in the Faculty of Science, Tokyo University.

I. Tomiyama (1936) figured two individuals of this goby as the blind forms of *Luciogobius guttatus guttatus* Gill. From the Tomiyama figures, C. T. Regan (1940) erected two new species, *Luciogobius pallidus* and *Luciogobius albus* separate from *Luciogobius guttatus* Gill.[4] In

[3] The author kept alive one of these blind gobies for 24 days, from 28th August, 1952, to 20th September. The goby was obtained in the cave in Daikonzima, Simane Pref., on 21st August, 1952. It was kept in a glass-jar at the laboratory, but was lost by careless management. During the period kept the water-temperature was from 22°C to 24°C, while at the natural habitat the temperature was 11.5°C. During that period the goby took no food, and became thinner day by day. It was about 55 mm in total length, and light pink in color. Generally no dark pigment appeared on the body, excepting a little which occasionally appeared on the posterior part of the body. From the observations upon the alive goby, it was supposed that the individual variation of body-form observed in the preserved specimens largely depended on the duration of the keeping-period after they were caught and the variation of pigmentation of body depended on the condition of pigmentation when they were thrown in the preservative.

[4] K. Matsubara (1955) accepted Regan's two species, *Luciogobius pallidus* Regan (Japanese name, Ido-mimizu-haze) and *Luciogobius albus* Regan (Japanese name, Dôkutu-mimizu-haze).

this study, the author reexamined fourteen specimens, which had been studied by I. Tomiyama (1936). The examined specimens exhibited remarkable individual variation of body-form in proportion of head and body-depth to length, pigmentation, forms of dorsal, ventral, anal and pectral fins, etc. (Table 1). C. T. Regan (1940) regarded these differences as the specific characteristics. But these differences appeared in each specimen in various degrees, so that the author could not recognize two separate species in those specimens as Regan did. From the reexamination of the preserved specimens and the observation of the above-mentioned alive fish (Foot Note 3), he came to the opinion that all these specimens belong to a single quite polymorphic, albinous *Luciogobius albus* Regan separate from *Luciogobius guttatus* Gill.

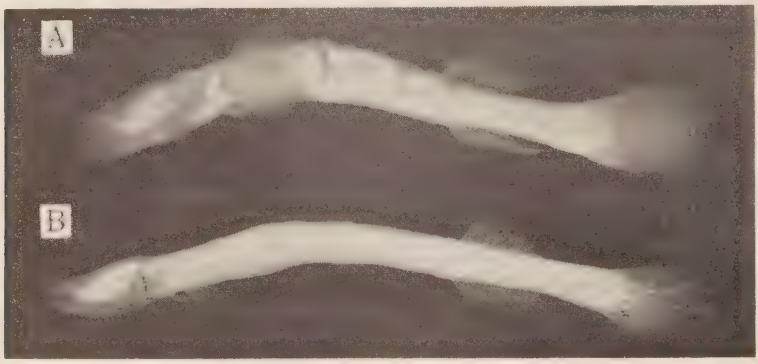

Fig. 2. *Luciogobius albus* Regan.

A, holotype, 43 mm in total length. B, one of the paratypes, 44 mm from Misaki, Kôti Pref.

Tomiyama's figures (1936) were drawn from these specimens, A and B. C. T. Regan (1940) erected *Luciogobius albus* from the figure of A and *L. pallidus* from the figure of B respectively.

B is damaged in anal, dorsal, and ventral fins.

Luciogobius elongatus Regan (Fig. 3)

(Japanese name, Naga-mimizu-haze)

Regan, C. T. 1905, 1940; Matsubara, K. 1955.

Synonym—*Luciogobius guttatus guttatus* Gill (part), Tomiyama, I. 1936.

Locality—Seto Inland Sea (Regan, C. T. 1905); Tanegasima, Kagosima Pref. (Snyder, J. O. 1912); Matugaura, Kagosima Pref. (The author also collected from Kagosima Pref.).

N. Kuroda (1953) recorded this goby as coming from Lake Biwa, the largest fresh-water lake in Japan, Siga Pref. But it seems to be erroneous as this goby has so far been collected only from the seashore.

C. T. Regan gave no figure of this species in his papers. The figure given here from a specimen, collected from Matugaura, is the first one published (Fig. 3).

Only one specimen, collected from a tide pool on the coast of Matugaura, was examined in this study.

Fig. 3. *Luciogobius elongatus* Regan.
40 mm in total length.

Expedio parvulus Snyder

(Japanese name, Nansen-haze)

Snyder, J. O. 1909; Matsubara, K. 1955.

Synonym—*Luciogobius guttatus parvulus* (Snyder), Tomiyama, I. 1936.

Locality—Misaki, Kanagawa Pref. (Snyder, J. O. 1909); Sirahana, Tiba Pref. (Sakamoto, K. 1932).

Three specimens, 37～48 mm in total length, were examined in this study. Those specimens had been studied by I. Tomiyama (1936).

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** original paper not referable to in this study.

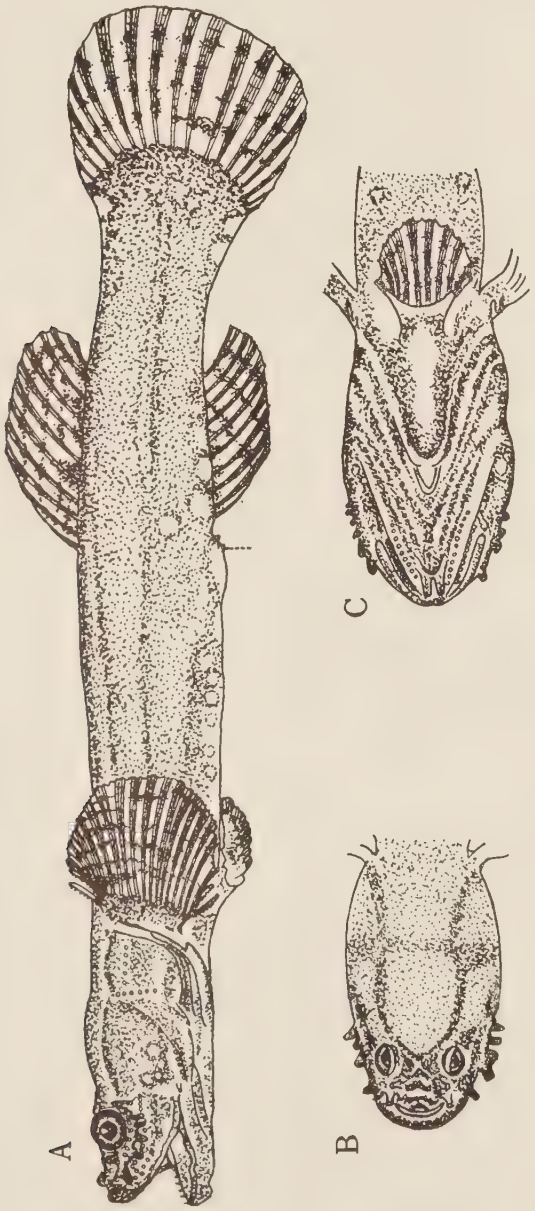
Explanation of Plate 2

Luciogobius saikaiensis n. sp.

Fig. A. Holotype, 41 mm in total length.

Fig. B. Dorsal side of A.

Fig. C. Ventral side of A.



Luciogobius saikaiensis n. sp.

Journal of the Faculty of Agriculture, Kyūshū University, Vol. 11, No. 1
August 30, 1957

On Copepoda of the family Corycaeidae in Japanese waters

Otohiko Tanaka

The species belonging to this family are, for the most part, high sea forms and are met with in the warm waters of oceans. The following six species had been known up until 1937 when Mori recorded fourteen species from the neighbouring waters of Japan. They are *C. speciosus* Dana, *C. robustus* Giesbrecht, *C. ovalis* Claus, *C. danae* Giesbrecht (*C. crassiusculus* Dana), *C. anglicus* Lubbock and *C. rostratus* Claus. Among these *anglicus* has been recorded from the European coast of the Atlantic, and the occurrence of this species in the Japanese waters cannot be accepted without further evidence; it is probably identical with the species described by Mori (1937) under the name *japonicus*. Mori recorded in 1937 the following fourteen species: *C. lautus* Dana, *C. speciosus* Dana, *C. crassiusculus* Dana, *C. agilis* Dana, *C. longistylis* Dana, *C. flaccus* Giesbrecht, *C. catus* F. Dahl, *C. latus* Dana, *C. ovalis* Claus, *C. asiaticus* F. Dahl, *C. trukicus* Mori, *C. gibbulus* Giesbrecht, *C. concinnus* Dana and *C. japonicus* Mori. The last species is, as pointed out by Davis (1949), identical with *C. affinis* McMurrich which is abundant in the northeastern Pacific at certain times of the year. The female specimen of *C. trukicus* is immature and doubtful; the male specimen of the species is, in my opinion, identical with the male of *C. andrewsi* Farran. Mori's *C. latus* and *C. catus* appear to be erroneous: the former is identical with *C. catus* F. Dahl, and the latter with *C. pacificus* F. Dahl.

I have collected twenty-three species from the warm waters of the Pacific coast and the Sea of Japan. They are: *C. speciosus*, *C. clausi* F. Dahl, *C. crassiusculus*, *C. viretus* Dana, *C. robustus* Giesbrecht, *C. typicus* Kröyer, *C. flaccus*, *C. limbatus* G. Brady, *C. longistylis*, *C. lautus*, *C. furcifer* Claus, *C. andrewsi* Farran, *C. asiaticus* F. Dahl, *C. subtilis* M. Dahl, *C. dubius* Farran, *C. dahli* nom nov., *C. affinis*, *C.*

agilis, *C. catus*, *C. pacificus* F. Dahl, *C. rostratus* Claus, *C. gibbulus* and *C. concinnus* Dana. Thus the species taken from the adjacent waters of Japan amount to 24 in number.

About fourty-one species of the family Corycaeidae have been reported from the oceans; the following thirteen species from both the Atlantic and Indo-Pacific: *C. rostratus*, *C. gracilis* Dana, *C. speciosus*, *C. clausi*, *C. flaccus*, *C. limbatus*, *C. lautus*, *C. furcifer*, *C. amazonicus* M. Dahl, *C. minimus* M. Dahl, *C. latus*, *C. giesbrechti* F. Dahl and *C. ovalis* Claus; four species from the Atlantic: *C. elongatus* Claus, *C. africanus* F. Dahl, *C. anglicus* Lubbock and *C. huxleyi* Lubbock; the following twenty-four species appear to be of the Indo-Pacific: *C. carinatus* Giesbrecht, *C. brevis* Farran, *C. curtus* Farran, *C. gibbulus* Giesbrecht, *C. concinnus*, *C. crassiusculus*, *C. viretus* Dana, *C. robustus*, *C. typicus* M. Dahl (*C. alatus* Giesbrecht), *C. longistylis*, *C. lubbocki* Giesbrecht, *C. tenuis* Giesbrecht, *C. asiaticus*, *C. erythraeus* Cleve, *C. dubius* Farran, *C. andrewsi*, *C. subtilis* M. Dahl, *C. agilis*, *C. pacificus*, *C. catus*, *C. pumilus* M. Dahl, *C. medius* Gurney, *C. brehmi* Steuer and *C. affinis*.

It is interesting to compare the Japanese species with those taken from the other regions of the Pacific, and from the Indian Oceans. The following number of species have been recorded from these regions: eighteen species from the Ceylon Pearl Fishery Bank by Thomson and A. Scott (1903); nine species from Bombay by Cleve (1903); eleven species from the Maldive and Laccadive Archipelago by Wolfenden (1905); sixteen species from the Malay region by A. Scott (1909); twenty species from the Christmas Island, Indian Ocean by Farran (1911); twenty-seven species from the Indo-Pacific by M. Dahl (1912); four species from the Suez Canal by Gurney (1912); twenty-four species from the Great Barrier Reef by Farran (1936); eleven species from the Indian Ocean by Sewell (1947); twenty-one species chiefly from the Pacific Ocean by Wilson (1950); two species from the San Diego region by Esterly (1905); one species from the northeastern waters of the North Pacific by Davis (1949). Among these species the following twelve species have not been recorded from the Japanese waters: *C. erythraeus* Cleve, *C. amazonicus* F. Dahl, *C brehmi* Steuer, *C. minimus* F. Dahl (*C. venustus* Dana?), *C. medius* Gurney, *C. pumilus* M. Dahl, *C. curtus* Farran, *C. carinatus* Giesbrecht and *C. pellucidus* Dana (*C. gracilis* Dana?).

It is probable that intensive study of copepoda extends the knowledge on the geographical distribution of the species, but a list of species without description and figures will bring much confusion to the question, and it is never superfluous to give them again.

Description of the species

Genus *Corycaeus* Dana

Subgenus *Corycaeus* M. Dahl

Corycaeus (*Corycaeus*) *speciosus* Dana

Plate 3, figs. 1—6

Corycaeus speciosus Giesbrecht, 1892, p. 673, taf. 51, figs. 39, 40.
Corycaeus speciosus A. Scott, 1909, p. 251.
Corycaeus (*Corycaeus*) *speciosus* M. Dahl, 1912, p. 13, taf. 1, 2.
Corycaeus speciosus Farran, 1936, p. 134.
Corycaeus speciosus Mori, 1937, p. 133, pl. 72, figs. 9–15.
Corycaeus (*Corycaeus*) *speciosus* Sewell, 1947, p. 276.
Corycaeus speciosus Wilson, 1950, p. 196.

Female. Length, 1.86–1.97 mm. Head and 1st thoracic segment partially fused. Posterior corners of 3rd thoracic segment exceed the distal margin of genital segment. Cephalothorax and 2-jointed abdomen in the proportional lengths 14 : 9. Abdominal segments and furcal rami in the proportional lengths calculated in parts per 100 are 38 : 16 : 46. Anterior segment, the head and 1st thoracic segment, is about 3/5 as wide as long. Long and divergent furcal rami are the characteristic feature of the species.

Male. Length, 1.48–1.62 mm. Head and 1st thoracic segment fused. Proportional lengths of cephalothorax and abdomen 5 : 4, and those of abdominal segments and furcal rami 33 : 21 : 46. Anterior segment is about 1.8 times as long as wide. Genital segment about 3/5 as wide as long (14 : 9). Anal segment more than 2 times as wide as long, and is of the same width throughout its length. Furcal rami slender, about 12 times as long as wide. Posterior corners of 3rd thoracic segment exceed the middle of the genital segment.

The species is very common in Japanese waters. According to M. Dahl it is widely distributed in the warm regions of the Pacific, Indian and Atlantic Oceans.

Corycaeus (*Corycaeus*) *clausi* F. Dahl

Plate 3, figs. 7—11

Corycaeus ovalis Giesbrecht, 1892, p. 659, taf. 51, figs. 1-3.
Corycaeus (*Corycaeus*) *clausi* M. Dahl, 1912, p. 18, taf. 2, figs. 5-12.
Corycaeus clausi Wilson, 1950, p. 193.

Female. Length, 1.50 mm. Cephalothorax, 1.1 mm, abdomen, 0.44 mm. Head fused with 1st thoracic segment. Proportional lengths of abdominal segments and furcal rami 46 : 22 : 32. Anterior segment about 1.6 times as long as wide. Genital segment oval, about as wide

as long (22 : 23). Anal segment wider at the proximal than at the distal (15 : 11). Furcal rami more than 3 times as long as it is wide at the proximal. Posterior corners of 3rd thoracic segment extend to the two-thirds of the genital segment.

Male. Length, 1.37 mm. Cephalothorax, 0.85 mm., abdomen, 0.52 mm. Proportional lengths of abdominal segments and furcal rami 46 : 21 : 33. Genital segment about 2 times as long as wide (7 : 4). Anal segment is by one-fifth wider at the proximal than at the distal (10 : 8). Furcal rami about 7 times as long as it is wide at the proximal (20 : 3).

The species is very rare in Japanese waters. The spcies has been recorded from the temperate waters of the Atlantic, but rarely from the Indo-Pacific.

Corycaeus (*Corycaeus*) *crassiusculus* Dana

Plate 3, figs. 12—15

Corycaeus danae Giesbrecht, 1192, p. 660, taf. 51, figs., 59, 60.
Corycaeus (*Corycaeus*) *crassiusculus* M. Dahl, 1912, p 21, taf. 3, figs. 1–7.
Corycaeus crassiusculus Farran, 1936, p. 134.
Corycaeus crassiusculus Mori, 1937, p. 133, pl. 75, figs 1–5.
Corycaeus (*Corycaeus*) *crassiusculus* Sewell, 1947, p. 272, fig. 69, a–h.
Corycaeus crassiusculus Wilson, 1950, p. 193.

Female. Length, 1.44–1.57 mm. Head separates from 1st thoracic segment. Proportional lengths of cephalothorax and abdomen 21 : 11, and those of abdominal segments and furcal rami 39 : 24 : 37. Anterior segment is about 2 times as long as wide (17 : 9). Genital segment is about 1.3 times as long as wide. Anal segment little wider at the proximal than at the distal (8 : 7). Distal margin of the genital segment extends on the dorsal surface over the anal segment. Furcal rami about 6 times as long as it is wide at the proximal. Posterior corners of 3rd thoracic segment extend to the distal end of the genital segment.

Male. Length, 1.27–1.36 mm. Head fused with 1st thoracic segment. Proportional lengths of cephalothorax and abdomen 51 : 31, and those of abdominal segments and furcal rami 40 : 21 : 39. Anterior segment about 1.8 times as long as wide (18 : 9). Genital segment about 1.5 times as long as wide (26 : 17). Anal segment little wider at the proximal than at the distal (8 : 7), and 2 times as long as it is wide at the distal. Furcal rami about 8 times as long as it is wide at the proximal. Posterior corners of 3rd thoracic segment not so slender as in the male of *C. speciosus*, and extend to the middle of the genital segment.

The species is very common in Japanese waters. It is, according to M. Dahl, widely distributed in the Indo-Pacific Ocean and the Mediterranean Sea.

Corycaeus (*Corycaeus*) *viretus* Dana

Plate 4, figs. 1—4

Corycaeus (*Corycaeus*) *viretus* M. Dahl, 1912, p. 25, taf. 3, figs. 8–13.
Corycneus viretus Farran 1936, p. 134, fig. 28, a-d.
Corycaeus viretus Wilson, 1950, p. 197.

Male. Length, 1.55–1.60 mm. Body much more inflated than the foregoing species. Cephalothorax 1.5 times as long as abdomen (26 : 17). Proportional lengths of abdominal segments and furcal rami 55 : 15 : 30. Anal segment is wider at the proximal than at the distal (13 : 11), and is about as long as it is wide at the distal (10 : 11). Furcal rami 5 times as long as wide (19 : 4).

The species is rare in Japanese waters. Six male specimens were collected from the Pacific coast of Middle-Japan and from the Sea of Japan. It has been recorded from the Indian Ocean near the west coast of Sumatra.

Subgenus *Monocorycaeus* M. Dahl

Corycaeus (*Monocorycaeus*) *robustus* Giesbrecht

Plate 4, figs. 5—8

Corycaeus robustus Giesbrecht, 1892, p. 673, taf. 51, figs. 38, 42.
Corycaeus robustus A. Scott, 1909, p. 251.
Corycaeus (*Monocorycaeus*) *robustus* M. Dahl, 1912, p. 27, taf. 4, figs. 1-8.
Corycaeus robustus Farran, 1936, p. 135.
Corycaeus robustus Wilson, 1950, p. 196.

Female. Length, 2.11 mm. Cephalothorax, 1.38 mm, abdomen 0.73 mm. The species is easily distinguished from the other members of the genus by the robust cephalothorax and genital segment. Proportional lengths of abdominal segments and furcal rami 59 : 12 : 29. Genital segment 1.4 times as long as wide (10 : 7).

The species is very rare in Japanese waters. Only a single female specimen was collected from the Pacific coast. The species has been recorded from the tropical region of the Pacific.

Subgenus *Agetus* M. Dahl

Corycaeus (*Agetus*) *typicus* Kröyer

Plate 4, figs. 9—15

Corycaeus alatus Giesbrecht, 1892, p. 674, taf. 51, figs 8, 9.

Corycaeus (*Agetus*) *typicus* M. Dahl, 1912, p 31, taf. 4, figs. 9-14, taf. 5, figs. 1-3.
Corycaeus typicus Farran, 1936, p. 135.
Corycaeus typicus Wilson, 1959, p. 196.

Female. Length, 1.50–1.61 mm. Head separates from 1st thoracic segment. Cephalothorax and abdomen in the proportional lengths 2 : 1. Genital segment and furcal rami in the proportional lengths 55 : 45. Inner marginal furcal seta flanged, and longer than the furcal ramus itself. The peculiar form of 4th thoracic segment and abdomen separate the species from the other members of the genus.

Male. Length, 1.50–1.54 mm. Cephalothorax and abdomen in the proportional lengths 8 : 5. Anterior segment less than 2 times as long as wide (40 : 23). Abdominal segments and furcal rami in the proportional lengths 52 : 18 : 30. Genital segment about 1.5 times as long as wide (33 : 23). Anal segment little wider at the proximal than at the distal (9 : 8). Furcal rami more than 6 times as long as it is wide at the proximal (19 : 3). Terminal claw of 2nd antenna about 2 times as long as the outer-marginal one arising from the proximal of the apical joint. In the other specimen dissected the claw is much shorter (3 : 2), and the spine of 1st basal joint of 2nd antenna longer than that of 2nd basal joint (4 : 3), whereas they are of about the same lengths in the ordinary specimen.

The species is fairly common in Japanese waters. Ten females and five males were collected from the Pacific coast. It has been recorded from the Indo-Pacific, the tropical region of the Atlantic and the Mediterranean Sea.

Corycaeus (*Agetus*) *flaccus* Giesbrecht

Plate 5, figs. 1—7

Corycaeus flaccus Giesbrecht, 1892, p. 674, taf. 51, figs. 10, 11.
Corycaeys flaccus A. Scott, 1909, p. 248.
Corycaeus (*Agetus*) *flaccus* M. Dahl, 1912, p. 35, taf. 5, figs. 4–11.
Corycaeus flaccus Farran, 1936, p. 135.
Corycaeus flaccus Mori, 1937, p. 135, pl. 73, figs. 7–15.
Corycaeus flaccus Wilson, 1950, p. 193.

Female. Length, 1.56–1.67 mm. Head separates from 1st thoracic segment. Cephalothorax and abdomen in the proportional lengths 9 : 4. Anterior segment 1.8 times as long as wide (47 : 26). Abdominal segments and furcal rami in the proportional lengths 4 : 3. Genital segment 1.5 times as long as wide, has a small rounded protuberance on the mid-dorsal of the segment. Lateral margins of genital segment are furnished with short hairs. Inner marginal furcal seta is the longest and is flanged.

Male. Length 1.37–1.39 mm. Cephalothorax and abdomen in the

proportional lengths 5 : 3. Anterior segment 3/5 as wide as long. Abdominal segments and furcal rami in the proportional lengths 47 : 20 : 33. Genital segment 1.5 times as long as wide (29 : 19); lateral distal margins of the segment produced when viewed from the dorsal. Anal segment wider at the proximal than at the distal (9 : 8). Furcal rami 7 times as long as wide (20 : 3). Terminal claw of 2nd antenna about 2.5 times as long as the outer-marginal one arising from the proximal of the apical joint, and about as long as the spines of the basal joints. Inner margin of 2nd basal joint has no large tooth, but is finely serrated.

The species is rather rare in Japanese waters. Seven females and three males were collected from the Pacific coast, and one male from the Japan Sea. The species is distributed in the warm waters of the Atlantic, especially in the Sargassum Sea, Mediterranean and Indo-Pacific.

Corycaeus (*Agetus*) *limbatus* G. Brady

Plate 5, figs. 8—12

Corycaeus elongatus Giesbrecht, 1892, p 674, taf. 51, fig. 6.
Corycaeus (*Agetus*) *limbatus* M. Dahl, 1912, p 38, taf. 5, figs. 12–14, taf. 6, figs. 1–5.
Corycaeus limbatus Farran, 1936, p. 136.
Corycaeus limbatus Wilson, 1950 p. 194.

Female. Length, 1.36 mm. Cephalothorax, 1.00 mm., abdomen, 0.36 mm. One-jointed abdomen is 2 times as long as furcal rami. The rhomboidal form of genital segment in dorsal aspect is a good specific character of the female.

Male. Length, 1.22 mm. Cephalothorax, 0.77 mm, abdomen, 0.45 Abdominal segments and furcal rami in the proportional lengths 52 : 17 : 29. Anterior segment less than 2 times as long as wide (7 : 4). The large cuticular lenses are contiguous. Genital segment about 1.6 times as long as wide (27 : 17). Anal segment is of the same width throughout its length, and 1.5 times as long as wide. Furcal rami 5 times as long as wide. Terminal claw of 2nd antenna about 2.5 times as long as the outer-marginal one.

The species is rare in Japanese waters. Three females and one male were collected from the Pacific coast. The species is distributed in the warm waters of the Atlantic and Indo-Pacific.

Subgenus *Urocorycaeus* M. Dahl

Corycaeus (*Urocorycaeus*) *longistylis* Dana

Plate 5, figs. 13—16

Corycaeus longistylis Giesbrecht, 1892, p. 674, taf. 51, fig. 37

Corycaeus (*Urocorycaeus*) *longistylis* M. Dahl, 1912, p. 42, taf. 6, 7.
Corycaeus longistylis A. Scott, 1909, p. 249.
Corycaeus longistylis Farran, 1936, p. 136.
Corycaeus longistylis Mori, 1937, p. 134, pl. 73, figs. 3–8.
Corycaeus (*Urocorycaeus*) *longistylis* Sewell, 1949, p. 277.
Corycaeus longistylis Wilson, 1950, p. 195.

Female. Length, 2.80 mm. Head fused with 1st thoracic segment. Third and fourth thoracic segments completely fused. Cephalothorax little longer than the combined length of abdominal segments and furca. Lateral corners of 3rd thoracic segment produced posteriorly to about the distal 1/3 of genital segment. Abdominal segments and furcal rami in the proportional lengths 21 : 21 : 58. Dorsal surface of genital segment vaulted. Furcal ramus slender and long, about 16 times as long as it is wide at the proximal.

Male. Length, 2.13–2.19 mm. Cephalothorax and abdomen in the proportional lengths 31 : 29. Abdominal segment and furcal rami are in the proportional lengths 13 : 18. Terminal claw of 2nd antenna about 3 times as long as the proximal outer-marginal one. There is a row of small teeth on the outer margin and the median line of 2nd basal joint of 2nd antenna.

The species is fairly common in Japanese waters. One female and fourteen males were collected from the Pacific coast. The species has been recorded only from the Pacific. Mori recorded the species from the southern waters of Japan.

Corycaeus (*Urocorycaeus*) *lautus* Dana

Plate 6, figs. 1—2

Corycaeus (*Urocorycaeus*) *lautus* M. Dahl, 1912, p. 45, taf. 7, figs. 4–14.
Corycaeus lautus Farran, 1936, p. 136.
Corycaeus lautus Mori, 1937, p. 132, pl. 72, figs. 1–8.
Corycaeus lautus Wilson, 1950, 194.

Male. Length, 2.29–2.32. Cephalothorax and abdomen in the proportional length 31 : 33. Abdominal segments and furcal rami in the proportional lengths 25 : 18 : 57. Proximal tooth on the inner distal margin of 2nd basal joint of 2nd antenna very sharp. Process of 2nd basal joint of 4th leg is furnished, beside a normal long seta, with a short one.

The species is rather rare in Japanese waters. Four males were collected from the Pacific coast. It has been recorded from the Sargassum Sea, the adjacent sea of New Guinea, and Pacific Ocean (113°W 13°S). Mori collected the species from the southern warm waters of Japan.

Corycaeus (*Urocorycaeus*) *furcifer* Claus

Plate 6, figs. 3—6

Corycaeus furcifer Giesbrecht, 1892, p. 674, taf. 51, figs. 41, 44-46.
Corycaeus furcifer A. Scott, 1909, p. 248.
Corycaeus (*Urocorycaeus*) *furcifer* M Dahl, 1912, p 48, taf. 8, figs. 1-7.
Corycaeus furcifer Farran, 1936, p. 136.
Corycaeus furcifer Wilson, 1950, p. 193.

Female. Length, 19.2 mm. Cephalothorax, 1.02 mm, abdomen, 0.90 mm. Abdominal segments and furcal rami in the proportional lengths 20 : 12 : 68. Lateral processes of 3rd thoracic segment extend to the proximal 1/3 of genital segment. Genital segment has a rounded process on the dorsal surface about the middle. Anal segment has two sharp spines on the dorsal surface at the distal margin.

The species is very rare in Japanese waters. A single female specimen was collected from the Pacific coast. It has been recorded from the Indo-Pacific, northern coast of South America, Mediterranean and Sargassum Seas.

Subgenus *Ditrichocorycaeus* M. Dahl

The species of this subgenus are, as Sewell (1947) pointed out, the most difficult of all copepods to discriminate. According to the key of the species given by M. Dahl they are divided into two. The first group has short furcal rami which are about as long as anal segment but shorter than genital segment; to this group belong *C. andrewsi*, *C. asiaticus* and *C. subtilis* M. Dahl. The second group is characterized by the possession of furcal rami which are longer than both anal segment and genital segment; to this group belong *C. amazonicus* F. Dahl, *C. anglicus* Lubbock, *C. minimus* M. Dahl, *C. tenuis* Giesbrecht, *C. lubbocki* Giesbrecht, *C. africanus* F. Dahl, *C. dubuis* Farran, *C. tenuis* Farran, *C. erythraeus* Cleve and *C. lubbocki* M. Dahl. Among those *C. lubbocki* Giesbrecht, *C. tenuis* Giesbrecht and *C. minimus* have no ventral hook on the genital segment in the female. M. Dahl (1912) described from the material taken in Sansibar, Ceylon and the China Sea, the species which had a ventral hook under the name *lubbocki*, and concluded that *C. tenuis* Farran is identical with her *lubbocki*. However, Sewell disagreed with M. Dahl's opinion and states that Giesbrecht's *lubbocki* which has no ventral hook is not identical with Dahl's *lubbocki*, and the true *lubbocki* Giesbrecht was redescribed in 1924 by Früchtl under the name *farrani*. He further states that *tenuis* Farran (non Giesbrecht) and *lubbocki* M. Dahl (non Giesbrecht) are probably synonymous, and both of them are synonymous of *C. africanus* F. Dahl. I have now three species of *Ditrichocorycaeus* belonging to

the second group; one of them is, without doubt, identical with *tenuis* Farran; the other two appear to be *dubius* Farran and *japonicus* Mori respectively. The last species is one of the most common one in the warm waters of Japan, and is synonymous, as indicated by Chales C. Davis (1949), with *C. affinis* McMurrich.

Corycaeus (*Ditrichocorycaeus*) *andrewsi* Farran

Plate 6, figs. 7—12

Corycaeus andrewsi Farran, 1911, p. 294, pl. 13, 14.
Corycaeus (*Ditrichocorycaeus*) *andrewsi* M. Dahl, 1912, p. 78, taf. 9, figs. 10–18.
Corycaeus andrewsi Farran, 1936, p. 138
Corycaeus trukicus Mori, 1937, p. 137, pl. 75, figs. 9–16.

Female. Length, 1.00–1.07 mm. Cephalothorax and abdomen in the proportional lengths 2 : 1. Head separates from 1st thoracic segment. Distal corners of 3rd thoracic segment produced into small wing-like expansions. Distal corners of 4th thoracic segment not extending to the middle of genital segment. Abdominal segments and furcal rami in the proportional lengths 47 : 29 : 24. There are two groups of fine hairs on the ventral surface of genital segment when viewed from the lateral, but no ventral hook. Inner marginal spine of 2nd basal joint of 2nd antenna about 3/5 times as long as that of 1st joint.

Male. Length, 0.82–0.88. Cephalothorax and abdomen in the proportional lengths 29 : 19. Abdomal segments and furcal rami in the proportional lengths 58 : 21 : 21. Anterior segment 1.5 times as long as wide (42 : 27). 2nd thoracic segment has on the lateral margins each a slight swelling about the middle. Genital segment 4/5 as wide as long, broadish oval in shape, and has a small ventral hook. Anal segment is of about the same width throughout its length, and 1.5 times as long as wide. Furcal rami 4 times as long as it is wide at the proximal. Wing-shaped expansions of 3rd thoracic segment extend to proximal 1/3 of genital segment. There is a longitudinal median row of spinules on 2nd basal joint of 2nd antenna.

Remarks. *Corycaeus trukicus* Mori belongs to the subgenus *Ditrichocorycaeus*. His female specimen has coarsely plumosed spines on each of the 1st and 2nd basal joints of 2nd antenna; this is a characteristic feature of the immature specimen of *Corycaeus* (sensu lato) except the subgenus *Corycella* Farran. *C. trukicus* is closely allied to *C. andrewsi*, *C. asiaticus*, and *C. subtilis* in having short furcal rami which are about as long as anal segment. His specimens measured 0.65–0.95 mm in female, and 0.8 mm in male. The female specimens deviate too much in total length. The smallest species among *Ditrichocorycaeus* is, according to M. Dahl, *C. subtilis* M. Dahl which measures 0.75–0.77 mm in female. *C. trukicus* differs from *C.*

subtilis in having a prolonged 4th thoracic segment, and a long plumose spine on the 2nd basal joint of 2nd antenna. The male specimen of *C. trukicus* agrees fairly well with the male of *C. andrewsi* in having furcal rami which are about as long as anal segment, and in having short wing-like expansions on the 3rd thoracic segment. My male specimen has on the lateral margins of 2nd thoracic segment a slight swelling on each side about the middle. This is clearly illustrated by Mori in his figure of the male specimen of *trukicus*.

This is one of the most common species in Japanese waters. Ten females and sixteen males were collected from the Pacific coast. The species has been recorded from the Indo-Pacific Ocean. Mori recorded the occurrence of the species from the station near Truk Island.

Corycaeus (*Ditrichocorycaeus*) *asiaticus* F. Dahl

Plate 6, figs. 13—19

Corycaeus murrayi Farran, 1911, p. 294, pl. 13, figs. 1–6.
Corycaeus (*Ditrichocorycaeus*) *asiaticus* M. Dahl, 1912, p. 74, taf. 11, figs. 1–9.
Corycaeus asiaticus Gurney, 1926, p. 163 fig. 24, a–c.
Corycaeus asiaticus Farran, 1936, p. 137.
Corycaeus asiaticus Mori, 1937, p. 136, pl. 75, figs. 6–8.
Corycaeus (*Ditrichocorycaeus*) *asiaticus* Sewell, 1947, p. 281.

Female. Length, 1.15–1.19 mm. Cephalothorax and abdomen in the proportional lengths 15 : 8. Anterior segment 1.6 times as long as wide. Abdominal segments and furcal rami in the proportional lengths 42 : 29 : 29. Genital segment about as long as wide (20 : 28), has no ventral hook; proximal ventral corner of the segment rectangular. Anal segment about as long as it is wide at the proximal (14 : 12); the proximal margin of segment 1.5 times as wide as it is wide at the proximal (12 : 8). Furcal rami 5 times as long as wide (14 : 3). Distal margin of 2nd basal joint of 2nd antenna has a small tooth distal to the large one. Apical spine of exopodite of 2nd leg is curved and without peculiar serration.

Male. Length, 1.04–1.12 mm. Cephalothorax and abdomen in the proportional lengths 3 : 2. Anterior segment 1.6 times as long as wide (14 : 9). Wing-like expansions of 3rd thoracic segment extend posteriorly about to the middle of genital segment. Abdominal segments and furcal rami in the proportional lengths 49 : 24 : 27. Genital segment 1.3 times as long as wide (23 : 18). Anal segment is of the same width throughout its length, and about 2 times as long as it is wide at proximal. Furcal rami about 6 times as long as it is wide at the middle of the ramus (25 : 4). Ventral hook of genital segment shows some individual difference.

The species is rather rare in Japanese waters. Four females and

three males were collected from the Pacific coast and one female from the Sea of Japan. It has been recorded from the Indo-Pacific.

Corycaeus (*Ditrichocorycaeus*) *subtilis* M. Dahl

Plate 7, figs. 1—4

Corycaeus (*Ditrichocorycaeus*) *subtilis* M. Dahl, 1912, p. 80, taf. 8, figs. 9–16.
Corycaeus subtilis Farran, 1936, p. 138.
Corycaeus subtilis Wilson, 1950, p. 196.

Female. Length, 0.76 mm. Cephalothorax, 0.52 mm, abdomen, 0.24 mm. Abdominal segments and furcal rami in the proportional lengths 44 : 26 : 30. Wing-like expansions of 3rd thoracic segment not extending to the middle of genital segment. Processes of posterior corners of 4th thoracic segment very small. Apical spine of exopodite of 2nd leg has two teeth on the inner anterior side.

The species is very rare in Japanese waters. Two females were collected from the Pacific coast. It has been recorded from the Indo-Pacific.

Corycaeus (*Ditrichocorycaeus*) *dubius* Farran

Plate 7, figs. 5—13

Corycaeus dubius Farran, 1911, p. 292, pl. 12, 14.
Corycaeus (*Ditrichocorycaeus*) *dubius* M. Dahl, 1912, p. 71, taf. 10, figs. 11–19.
Corycaeus erythraeus Gurney, 1926, p. 161, fig. 23, a–d.
Corycaeus erythraeus Farran, 1936, p. 137.
Corycaeus dubius Wilson, p. 193.

Female. Length, 109 mm. Cephalothorax, 0.65 mm, abdomen, 0.44 mm, so the abdomen is contained about 5 times in the length of the cephalothorax. Cephalothorax rather slender, about 2.4 times as long as wide (90 : 38). Head separates from thoracic segment. Posterior corners of 3rd thoracic segment exceed slightly beyond the middle of genital segment. 4th thoracic segment short, with sharp points. Proportional lengths of abdominal segments and furca measured ventrally 31 : 33 : 36; another specimen measuring 1.04 mm in overall length has the proportional length 29 : 35 : 36. Genital segment about 1.3 times as long as wide with a sharp hook directing posteriorly at the proximal corner of the segment when viewed from the lateral. The sides of 2nd abdominal segment parallel, about 2.5 times as long as wide. Furcal rami slightly divergent, 10 times as long as wide. The spine arising from 2nd basal joint of 2nd antenna slightly less than 1/3 the length of that from 1st basal joint, reaches the middle of the joint. Apical spine of exopodite of 2nd leg has about 5 teeth on the inner anterior side.

Male. Length, 0.836 mm. Cephalothorax, 0.45 mm, abdomen, 0.386 mm, so the abdomen is contained 1.2 times in the length of cephalothorax. Posterior corners of 3rd thoracic segment do not extend to the middle of genital segment. Abdominal segments and furcal rami in the proportional lengths 39 : 26 : 35. This proportion differs slightly from those given by M. Dahl (40.3 : 24.2 : 35.5) or Gurney (41.8 : 24.0 : 34.2). Teeth on the inner distal margin of 2nd basal joint of 2nd antenna stronger than those figured by M. Dahl. The characteristic teeth on the inner anterior side of the apical spine of the exopodite of 2nd leg not observed in the male specimen.

The species is rare in Japanese waters. A single male from the Pacific coast and two females from the Sea of Japan. The species has been recorded from the tropical regions of the Pacific and Indian Oceans, and also from the Suez Canal.

Remarks. The present specimen, though slightly larger in size than those of Farran (female 0.97 mm, from Christmas Island; female 0.96–1.02 mm, male 0.84–0.91 mm from the Great Barrier Reef), agrees quite well with his description and figures of *C. dubius*. Gurney (1927, p. 161) states that his *erythraeus* collected from the Suez Canal are probably be identical with *erythraeus* Cleve taken from the Red Sea. Farran (1936, p. 137), regarding Gurney's identification as correct, is of opinion that his *dubius* is identical with *erythraeus* Cleve. I agree that *erythraeus* Gurney is a synonym of *dubius* Farran, as both specimens have the similar structure in the 2nd antenna. It seems probable that these species may be synonymous but one cannot prove it until the doubtful species, *erythraeus* Cleve will be more fully described.

Corycaeus (*Ditrichocorycaeus*) *dahli* sp. nov.

Plate 7, figs. 14—17, Plate 8, figs. 1—5

Corycaeus (*Ditrichocorycaeus*) *lubbocki* M. Dahl, 1912, p. 64, taf. 10, figs. 20–28.
Corycaeus tenuis Farran, 1911, p. 291, pl. 12, figs. 8–9.
Corycaeus (*Ditrichocorycaeus*) *africanus* Sewell, 1947, p. 279, text-fig. 70, a–d.

Female. Length, 1.03–1.08 mm. Cephalothorax about 1.7 times as long as abdomen (68 : 40). Abdominal segments and furcal rami in the proportional lengths 35 : 23 : 42. Anterior segment 1.83 time as long as wide (53 : 29). Posterior corners of 3rd thoracic segment extend beyond the middle of genital segment. Fourth thoracic segment produced into short pointed processes. Genital segment about 1.3 times as long as wide; dorsal surface of the segment vaulted; there is a folding on the distal 2/3 of the dorsal surface; the ventral surface rather flat. Anal segment wider at the proximal than at the distal, and is 2.6 times as long as it is wide at the distal. Furca divergent,

about 16 times as long as it is wide at the distal. The spine arising from 1st basal joint of 2nd antenna 2 times as long as that from 2nd basal. Apical spine of exopodite of 2nd leg curved, and has about 6 teeth on the inner anterior side.

Male. Length, 0.89–0.91 mm. Abdomen is contained 1.4 times in the length of cephalothorax (57 : 41). Cephalothorax 1.5 times as long as wide (19 : 12.5). Propotional lengths of abdominal segments and furcal rami 50 : 18 : 32. Genital segment oval in shape, 1.6 times as long as wide (35 : 21); ventral surface of the segment has a small median hook; cylindrical part of the segment about 1.5 times as long as it is wide at the proximal (14 : 9). Furcal rami 8 times as long as wide (25 : 3); the longest furcal seta longer than the furcal ramus itself. In 2nd antenna terminal claw is longer than 2nd basal joint, and 3 times as long as proximal hook of terminal joints; spine arising from 1st basal joint of 2nd antenna little longer than that of 2nd joint. Apical spines of exopodite of the first three legs are straight. Outer-edge spine of 3rd joint of exopodite of 4th leg longer than half the length of 3rd joint (3 : 4); outeredge spine of 1st joint of exopodite about half as long as 1st joint of the same leg.

Remark. Nine species of *Ditrichocorycaeus* have been reported which resemble each other so closely that considerable confusion has arisen in regarding the species. They are *C. lubbocki* Giesbrecht, *C. lubbocki* M. Dahl, *C. lubbocki* Sewell, *C. tenuis* Giesbrecht, *C. tenuis* Farran, *C. tenuis* M. Dahl, *C. africanus* F. Dahl, *C. africanus* Sewell and *C. brehmi* Steuer. Proportional lengths of abdominal segments and furca of the species in parts per 100 as follows:

	Total length mm.	Genital	2nd	Furcal ramus
C. lubbocki Giesbrecht	0.95	37	17	46
C. lubbocki Sewell	1.033	37.2	15.2	47.6
C. lubbocki M. Dahl	0.95—0.97	35.2	22.5	42.3
C. tenuis Giesbrecht	0.87	37	21	42
C. tenuis Farran	1.05	35	20	45
C. tenuis M. Dahl	0.81—0.84	34.5	22.4	43.1
C. africanus M. Dahl	1.01—1.03	37.5	18.75	43.75
C. africanus Sewell	—	35.5	22.1	42.4
C. brehmi	0.95—1.10	41.8	18.6	39.6
Present specimens	1.03—1.08	35	23	42

Putting aside *lubbocki* Giesbrecht, *tenuis* Giesbrecht and *lubbocki* Sewell which have no ventral hook, and *C. brehmi*, the remaining five have no remarkable difference between them. It is clear that *C. lubbocki* M. Dahl is, according to the description and figures given by both

Giesbrecht and Sewell (1947, p. 281), not identical with *C. lubbocki* Giesbrecht. M. Dahl's *lubbocki* is, as she states, the synonym of *C. tenuis* Farran. Sewell described a form which was taken from the Indian Ocean, and referred it to *C. africanus* F. Dahl. However, his specimen is more closely allied to *C. lubbocki* M. Dahl than to *africanus* in having the abdominal segments and furcal rami which are nearer in the proportional lengths to those of *lubbocki* M. Dahl. Moreover, the spine of 1st basal joint of 2nd antenna is, according to Sewell's figure, less than 3 times the length of that of the 2nd basal joint, whereas it is 3 times in *africanus*. It is probable that *lubbocki* M. Dahl and *tenuis* Farran are, as M. Dahl pointed out, synonymous. Sewell is of opinion that both *tenuis* Farran and *lubbocki* M. Dahl are synonyms of *africanus*, but it seems to me that he is not correct. *C. africanus* differs from those two forms not only in the proportional lengths of the abdomen, but also in its geographical distribution. Sewell's record of *africanus* from the Indo-Pacific seems to be a conjecture. He was forced to choose one of these species, *tenuis*, *lubbocki* and *africanus*. Consequently, he picked up *africanus* which had been recorded from the west coast of Africa, and referred his specimen to it. His single specimen agrees, as he states, with *tenuis* Farran. The present specimen comes near to *lubbocki* M. Dahl, *tenuis* Farran and *africanus* Sewell in the proportional lengths of abdominal segments and furca, also in the total lengths. But I am not sure that the specimen is quite identical with the latter three species, as no mention has been made by the authors on the possession of the serration along the inner anterior side of the apical spine of exopodite of 2nd leg. This serration is apt to be overlooked when it is wrongly situated under microscope. For instance, two figures of the same apical spine viewed in two different aspects are given (plate V, fig. 17); it is easily overlooked when laid as shown in fig. 17, left. I have detected such serration in *C. dubius* Farran, the present specimen and *C. affinis*. The serration appears to be a characteristic feature of some of the female specimens of *Ditrichocorycaeus*. In my opinion, *lubbocki* M. Dahl, *tenuis* Farran and *africanus* Sewell and the present specimen are synonymous. Here I propose, to avoid further confusion, and to give a new name *dahli* to the specimen which has the characteristics as shown in the descriptive note. I wished the name *farrani*, but the name had been reoccupied by Früchtl. Dr. A. Fleminger of Harvard University has recently sent me specimens of *Corycaeus* (*Ditrichocorycaeus*) *americanus* taken from the Gulf of Mexico which appears to be new to science. The female specimen have total lengths 1.12–1.31 mm; abdominal segments and furcal rami in the proportional lengths 28 : 15 : 57. The species is characterized by its long furcal rami. The male specimen measured 0.90–1.01 mm in total length.

The species is rather rare in Japanese waters. Four females and eight males were collected from the Sea of Japan, and three males from the Pacific coast. It has been recorded from the tropical regions of the Pacific and Indian Oceans, also from the Great Barrier Reef Sea.

Corycaeus (*Ditrichocorycaeus*) *affinis* McMurrich

Plate 8, figs. 6—15

Corycaeus japonicus Mori, 1937. p. 138, pl. 76, figs. 1–11.
Corycaeus affinis Davis, 1949, p, 75, figs. 179–183.

Female. Length, 1.01–1.25 mm. Proportional lengths of cephalothorax and abdomen 77 : 43, and those of abdominal segments and furcal rami 36 : 26 : 38. Anterior segment 1.8 times as long as wide (31 : 27). Genital segment 3/4 as wide as long. Anal segment little wider at the proximal, and less than 2 times as long as it is wide at the distal (11 : 6). Furcal rami 1.7 times as long as anal segment (17 : 10), and about 9 times as long as at is wide at the proximal (17 : 2); the rami divergent. Dorsal surface of genital segment vaulted, and the vaulted portion is slightly folded between the genital flaps; ventral surface of genital segment produced prominently below at the middle. In 2nd antenna 2nd basal joint 2 times as long as wide; spine arising from 1st basal joint 3 times as long as that from 2nd basal. Apical spine of exopodite of 2nd leg rather straight, has two or sometimes three teeth on the inner anterior side. Outer-edge spine of 3rd joint of exopodite of 4th leg longer than half the length of the joint itself; outer edge spine of 1st joint of exopodite shorter than half the length of the joint.

Male. Length, 0.83 mm. Cephalothorax 1.6 times as long as abdomen (51 : 38). Proportional lengths of abdominal segments and furcal rami 50 : 22 : 28. Eyes separated by about 1/3 the greatest diameter. Anterior segment about half the total length of body, 3/5 as wide as long (31 : 50). 3rd thoracic segment has sharp backward pointing wings which extend to proximal 1/3 of the genital segment. The demarcation between 3rd and 4th thoracic segments indistinct. 4th thoracic segment produced into small pointed processes. Genital segment 1.7 times as long as wide (20 : 12); ventral surface of the segment furnished with a ventrally pointing hook; cylindrical part of the segment about 1/3 the length of the distal margin of the segment. Anal segment 1.6 times as long as it is wide at the distal (8 : 5). Furcal rami straight, are of about equal width throughout its length; they are about 6 times as long as wide (11 : 2); the longest seta is about twice as long as furcal ramus (19 : 11). Inner margin of 2nd basal joint of 2nd antenna furnished with two teeth, one of which very strong; outer

margin of 2nd basal joint is furnished with fine teeth; the anterior surface is also furnished with a row of very fine teeth. Spine arising from 1st basal joint of 2nd antenna 5/4 as long as that from 2nd basal which is furnished with fine hairs. Apical spine of exopodite of 1st leg curved outwardly; that of 2nd leg also curved, and is shorter than that of 3rd leg. Apical spine of exopodite of 3rd leg is straight, and is longer than the 3rd joint of exopodite. 4th leg has two setae on the process of 2nd basal joint; outer-edge spine of 1st joint of exopodite 2/3 the length of the 1st joint, and shorter than that of 3rd joint of exopodite which is about as long as the 3rd joint itself.

Remarks. The present female specimen is closely allied to the foregoing species in its general appearance. However, it differs from *dahli* in the proportional lengths of abdominal segments and furcal rami, namely furcal rami are much shorter in the present specimen. Apical spine of expodite of 2nd leg is straight, and the teeth of inner anterior side is less in number. The male specimen has the abdomen which differs also in the proportional lengths from those of *dahli*.

C. Davis states that *japonicus* Mori is identical with *affinis* McMurrich (1916) which is abundant in the waters near Friday Harbor, Washington at certain times of the year. According to his description the proportional lengths of abdominal segment and furcal rami of female are 10 : 6 : 10 (38.4 : 23.2 : 38.4 = 100); genital segment is as long as furcal ramus. Mori does not mention the proportional lengths of abdominal segments and furca of the female but his figure illustrates that the proportional lengths measured dorsally are 38 : 24 : 38, which come very near to my measurements; the genital segment has a folding on the dorsal surface. Thus these characteristics of a female specimen of *japonicus* agree well with those of *C. affinis* described by Davis. The male specimen of *japonicus* has the abdomen quite similar in proportional lengths to those of *affinis*. Mori's specimens measured about 1.1 mm in female, and 0.9 mm in male. Davis's measured 1.1 mm in female, 0.82 0.9 mm in male. *C. anglicus* Lubbock which has been recorded by some of the Japanese authors from the Japanese waters is probably identical with the present species.

The species is the most common one in the warm waters of Japan. It appears to have a wide distribution in the North Pacific, and has been recorded from the east and west coasts of Vancouver Island, Friday Harbor, Washington. It has been recorded by Mori from southern waters of the Pacific coast, and from the Inland Sea, and also from the Sea of Japan. I have detected the species from both the Pacific coast and from the Sea of Japan.

Subgenus *Onychocorycaeus* M. Dahl

Corycaeus (*Onychocorycaeus*) *agilis* Dana

Plate 8, figs. 16—18, Plate 9, figs. 1—5

Corycaeus gracilicaudatus Giesbrecht, 1892, p. 674, taf. 51, figs. 15, 30.
Corycaeus (*Onychocorycaeus*) *agilis* M. Dahl, 1912, p. 84, taf. 12.
Corycaeus agilis Farran, 1936, p. 138.
Corycaeus agilis Mori, 1937, p. 72, figs. 1–2.
Corycaeus (*Onychocorycaeus*) *agilis* Sewell, 1947, p. 284.
Corycaeus agilis Wilson, 1950, p. 192.

Female. Length, 0.88–1.00 mm. Cephalothorax robust, 1.5 times as long as abdomen. Abdominal segments and furcal rami slender, has proportional lengths 35 : 28 : 36.

Male. Length, 0.71–0.74 mm. Cephalothorax and abdomen in proportional lengths 47 : 36. Abdominal segments and furcal rami in proportional lengths 40 : 27 : 33. Genital segment is in the oval part 3/4 as wide as long. Some specimens had each a minute median hook on the ventral proximal corner of genital segment. The specimen from the Japan Sea measured 1.04 mm in female, and 0.79–0.81 mm in male.

The species is widely distributed in the warm waters of Japan, and has been recorded from the tropical regions of the Atlantic, Pacific and Indian Oceans.

Corycaeus (*Onychocorycaeus*) *catus* F. Dahl

Plate 9, figs. 6–12

Corycaeus obtusus Giesbrecht, 1892, p. 673, taf. 51, figs. 12–14.
Corycaeus catus Farran, 1911, p. 290, pl. 12, figs. 1–3.
Corycaeus (*Onychocosycaeus*) *catus* M. Dahl, 1912, p. 99, taf. 13, figs. 17–24.
Corycaeus catus Farran, 1936, p. 138.
Corycaeus latus Mori, 1937, p. 136, pl. 74, figs. 8–10.
Corycaeus (*Onychorycaeus*) *catus* Sewell, 1947, p. 284.
Corycaeus catus Wilson, 1950, p. 192.

Female. Length, 0.93–1.00 mm. Body robust. Abdomen is contained more than 2 times in the length of cephalothorax (9 : 4). Abdominal segments and furcal rami in the proportional lengths 58:20:22. Anal segment shorter than it is wide at the proximal (7 : 8). Furcal rami about 4 times as long as wide (14 : 4). Outer-edge spine of 1st joint of exopodite of 4th leg long, and exceeds the proximal end of 2nd joint; outer edge spine of 3rd joint is about as long as the 3rd joint of exopodite.

Male. Length, 0.80–0.90 mm. Head separates from 1st thoracic segment. Abdomen is contained 1.5 times in the length of cephalothorax (60 : 38). Abdominal segments and furcal rami in the proportional lengths 59 : 19 : 22. Genital segment has a ventral median hook.

Apical spine of exopodite of 2nd leg is straight but curved slightly inwards near the distal end, and the serrated outer margin is continued towards the inner margin near the apex.

Mori's *latus* is identical with the present species in having similar proportional lengths in abdominal segments and furcal rami, and the slender wing-like expansion of 4th thoracic segment in the female. His male specimen of *C. latus* has also general resemblance to the male specimen of *C. catus* F. Dahl in the total length and also in the proportional lengths of abdominal segments and furcal rami.

The species is very common in the warm waters of Japan. It has been recorded from the Indo-Pacific Ocean, Great Barrier Reef Sea, and Arabian Sea.

Corycaeus (*Onychocorycaeus*) *pacificus* F. Dahl

Plate 9, figs. 13–29

Corycaeus (*Onychocorycaeus*) *pacificus* M. Dahl, 1912, p. 103, taf. 14.
Corycaeus pacificus Farran, 1936, p. 139.
Corycaeus catus Mori, 1937, p. 74, figs. 1–7.
Corycaeus (*Onychocorycaeus*) *pacificus* Sewell, 1947, p. 285.
Corycaeus pacificus Wilson, 1950, p. 195.

Female. Length, 1.12–1.21 mm. Cephalothorax robust. Abdomen is contained about 2 times in the length of cephalothorax (88 : 42). Third thoracic segment is very wide; posterior corners of the segment extend to 3/4 of the genital segment. Wings of 4th thoracic segment short and bluntly pointed. Abdominal segments and furcal rami in the proportional lengths 56 : 22 : 22. Genital segment 1.4 times as long as wide (24 : 17). Anal segment wider at the proximal than at the distal (2 : 6), and as long as it is wide at the proximal (9 : 8). Furcal rami about 5 times as long as wide (9 : 2). Apical spine of exopodite of 2nd leg straight, and has an isolated tooth on the inner margin near the apical portion.

Male. Length, 0.99–1.09 mm. Abdomen is contained 1.2 times in the length of the cephalothorax (64 : 46). Abdominal segments and furcal rami in the proportional lengths 54 : 21 : 25. Genital segment oval, and 1.5 times as long as wide (26 : 17). Anal segment about 2 times as long as wide (11 : 6). Furcal rami 8 times as long as wide (12 : 1.5). Genital segment is devoid of ventral hook.

Mori's *catus* is clearly identical with *C. pacificus*; his female specimen of *catus* has narrowly rounded forehaed and large wing-like expansions on the 3rd thoracic segment extending beyond the middle of the genital segment. His male specimen of *catus* differs from mine in having a ventral hook on the genital segment, and also in the proportional lengths of the abdominal segments and furcal rami.

The species is very common in Japanese waters. The species has a wide distribution in the Pacific and Indian oceans.

Subgenus *Corycella* Farran

Corycaeus (*Corycella*) *rostratus* (Claus)

Corycaeus rostratus Giesbrecht, 1892, p. 674, taf. 5, 9.
Corycaeus rostratus Thompson and A. Scott, 1903, p. 285.
Corycaeus rostratus Steuer, 1910, p. 31.
Corycaeus (*Corycella*) *rostratus* M. Dahl, 1912, p. 111, taf. 5.

Male. Length, 0.69 mm. Cephalothorax, 0.44 mm, abdomen, 0.25 mm. Genital segment 1/3 as wide as long. Furcal rami 3 times as long as wide.

The species is very rare. A single male specimen was collected from the Pacific coast. The species is distributed in the subtropical regions of the Pacific and Atlantic Oceans, also in the Mediterranean Sea.

Corycaeus (*Corycella*) *gibbulus* (Giesbrecht)

Corycaeus gibbulus Giesbrecht, 1892, p. 675, taf. 51.
Corycaeus gibbulus Thompson and A. Scott, 1903, p. 286.
Corycaeus (*Corycella*) *gibbulus* M. Dahl, 1912, p. 115, taf. 15.
Corycella gibbula Farran, 1936, p. 139.
Corycaeus gibbulus Mori, 1937, p. 137, pl. 76, figs. 12–16, pl. 77, figs. 1–4.

Female. Length, 0.85–1.00 mm. Cephalothorax, 0.62–0.67 mm, abdomen, 0.27–0.30 mm. The species can be easily recognized by the peculiar form of genital segment.

Male. The male specimen of *C. gibbulus* and C. *concinnus* differ, as M. Dahl states, only in size. I follow Dahl's suggestion and decide the larger specimen (0.80–0.87 mm) as the male of *C. gibbulus*, and the small one (0.73–0.78 mm) as *C. concinnus*.

The species is very common in Japanese waters. It has a wide distribution in the warm regions of the Indian Ocean.

Corycaeus (*Corycella*) *concinnus* (Dana)

Corycaeus concinuus Giesbrecht, 1892, p. 675, taf. 51, figs. 21–24.
Corybaeus concinnus Thompson and A. Scott, 1903, p. 286.
Corycaeus concinnus A. Scott, 1909, p. 246.
Corycaeus (*Corycella*) *concinnus* M. Dahl, 1912, p. 121, taf. 15.
Corycella concinna Farran, 1936, p. 139.
Corycaeus concinnus Mori, 1937, p. 138, pl. 77, figs. 5–12.

Female. Length, 0.84–0.90 mm. The present specimen agrees quite well with Dahl's description.

The species is very common in Japanese waters. It is distributed in the subtropical region of the Indian and Pacific Oceans.

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Explanation of Plates

Plate 3.

Figs. 1—6. *Corycaeus* (*Corycaeus*) *speciosus* Dana

Fig. 1. Female, dorsal view. × 30
Fig. 2. Female, abdomen, lateral view. × 60
Fig. 3. Female, 2nd antenna. × 120
Fig, 4. Female, 4th leg. × 120
Fig. 5. Male, dorsal view. × 30
Fig. 6. Male, 2nd antenna. × 120

Figs. 7–11. *Corycaeus* (*Corycaeus*) *clausi* F. Dahl

Fig. 7. Female, dorsal view. × 30
Fig. 8. Female, abdomen, lateral view. × 60
Fig. 9. Female, 2nd antenna. × 173
Fig. 10. Female, 4th leg. × 173
Fig. 11. Male, dorsal view. × 30

Figs. 12—14. *Corycaeus* (*Corycaeus*) *crassiusculus* Dana

Fig. 12. Female, dorsal view. × 30
Fig. 13. Female, abdomen, lateral view. × 60
Fig. 14. Female, 2nd antenna. × 120
Fig. 15. Male, dotsal view. × 30

Plate 4.

Figs. 1—4. *Corycaeus* (*Corycaeus*) *viretus* Dana

Fig. 1. Male, dorsal view. × 30
Fig. 2. Male, lateral view. × 30
Fig. 3. Male, 2nd antenna. × 120
Fig. 4. Male, 4th leg. × 173

Figs. 5—8. *Corycaeus* (*Monocorycaeus*) *robustus* Giesbrecht

Fig. 5. Female, dorsal view. × 26
Fig. 6. Female, abdomen, lateral view. × 30
Fig. 7. Female, 2nd antenna. × 90
Fig. 8. Female, 4th leg. × 120

Figs. 9—15. *Corycaeus* (*Agetus*) *typicus* Kröyer

Fig. 9. Female, dorsal view. × 30
Fig. 10. Female, lateral view. × 30
Fig. 11. Female, 2nd antenna. × 120
Fig. 12. Female, 4th leg. × 173.
Fig. 13. Male, dorsal view. × 30
Fig. 14. Male, abdomen, lateral view. × 60
Fig. 15. Male, 2nd antenna, × 173

Plate 5.

Figs. 1—7. *Corycaeus* (*Agetus*) *flaccuss* Giesbrecht

Fig. 1. Female, lateral view. × 30
Fig. 2. Female, dorsal view. × 30
Fig. 3. Female, 2nd natenna. × 120
Fig. 4. Female, 4th leg. × 173
Fig. 5. Male, dorsal view. × 30
Fig. 6. Male, abdomen, lateral view. × 60
Fig. 7. Male, 2nd antenna. × 173

Fgs. 1—12. *Corycaeus* (*Agetus*) *limbatus* G. Brady

Fig. 8. Female, dorsal view. × 30
Fig. 9. Female, lateral view. × 30
Fig. 10. Female, 2nd antenna. × 120
Fig. 11. Male. dorsal view. × 45
Fig. 12. Male, lateral view. × 45

Figs. 13—16. *Corycaeus* (*Urocorycaeus*) *longistylis* Dana

Fig. 13. Female, dorsal view. × 30
Fig. 14. Male, dorsal view. × 30
Fig. 15. Male, 2nd antenna. × 120
Fig, 16. Male, 4th leg. × 120

Plate 6.

Figs. 1—12. *Corycaeus* (*Urocorycaeus*) *lautus* Dana

Fig. 1. Male, dorsal view. × 26
Fig. 2. Male, 4th leg. × 120

Figs. 3—6. *Corycaeus* (*Urocorycaeus*) *furcifer* Claus

Fig. 3. Female, dorsal view. × 30
Fig. 4. Female, abdomen, lateral view. × 120
Fig. 5. Female, 2nd antenna. × 120
Fig. 6. Female, 4th leg. × 173

Fig. 7—12. *Corycaeus* (*Ditrichocorycaeus*) *andrewsi* Farran

Fig. 7. Female, dorsal view. × 45
Fig. 8. Female, abdomen, lateral view. × 120
Fig. 9. Female, 2nd antenna. × 173
Fig. 10. Male, dorsal view. × 60
Fig. 11. Male, abdomen, lateral view. × 120
Fig. 12. Male, 2nd antenna. × 240

Figs. 13—19. *Corycaeus* (*Ditrichocorycaeus*) *asiaticus* F. Dahl

Fig. 13. Female, dorsal view. × 35
Fig. 14. Female, abdomen, lateral view. × 70
Fig. 15. Female, 2nd antenna. × 100
Fig. 16. Female, apical spine of exopodite of 2nd leg. × 140
Fig. 17. Female, 4th leg. × 150
Fig. 18. Male, dorsal view. × 50
Fig. 19. Male, abdomen, lateral view. × 100

Plate 7.

Figs. 1—4. *Corycaeus* (*Ditrichocorycaeus*) *subtilis* M. Dahl

Fig. 1. Female, lateral view. × 60
Fig. 2. Female, abdomen, dorsal view. × 120
Fig. 3. Female, 2nd antenna. × 240
Fig. 4. Female, apical spine of exopodite of 2nd leg. × 240

Figs. 5—13. *Corycaeus* (*Ditrichocorycaeus*) *dubius* Farran

Fig. 5. Female, dorsal view. × 50
Fig. 6. Female, abdomen, lateral view. × 50
Fig. 7. Female, 2nd antenna. × 200
Fig. 8. Female, apical spine of exopodite of 2nd leg. × 200
Fig. 9. Female, 4th leg. × 200
Fig. 10. Male, dorsal view. × 60
Fig. 11. Male, abdomen, lateral view. × 120
Fig. 12. Male, 2nd antenna. × 240
Fig. 13. Male, apical spine of exopodite of 1st leg. × 240

Figs. 14—17. *Corycaeus* (*Ditrichocorycaeus*) *dahli*, nom. nov.

Fig. 14. Female, lateral view. × 50
Fig. 15. Female, abdomen, dorsal view. × 70
Fig. 16. Female, 2nd antenna. × 200
Fig. 17. Female, apical spine of exopodite of 2nd leg viewed in two different aspects. × 200

Plate 8.

Figs. 1—5. *Corycaeus* (*Ditrichocorycaeus*) *dahli*, sp. nov.

Fig. 1. Male, dorsal view. × 60
Fig. 2. Male, abdomen, lateral view. × 120
Fig. 3. Male, 2nd antenna. × 240
Fig. 4. Male, apical spine of exopodite of 2nd leg. × 240

Figs. 6—15. *Corycaeus* (*Ditrichocorycaeus*) *affinis* McMurrich

Fig. 6. Female, dorsal view. × 60
Fig. 7. Female, abdomen, lateral view. × 120
Fig. 8. Female, 2nd antenna. × 240
Fig. 9. Female, apical spine of exopodite of 2nd leg. × 200
Fig. 10. Female, 4th leg. × 240
Fig. 11. Male, dorsal view. × 60
Fig. 12. Male, abdomen, lateral view. × 120
Fig. 13. Male, 2nd antenna. × 240
Fig. 14. Male, apical spine of exopodite of 1st leg. × 240
Fig. 15. Male, 4th leg. × 40

Figs. 16—18. *Corycaeus* (*Onychocorycaeus*) *agilis* Dana

Fig. 16. Female, dorsal view. × 60
Fig. 17. Female, abdomen, lateral view. × 60
Fig. 18. Female, 4th leg. × 240

Plate 9.

Figs. 1—5. *Corycaeus* (*Onychocorycaeus*) *agilis* Dana

Fig. 1. Female, 2nd antenna. × 173
Fig. 2. Male, dorsal view. × 60
Fig. 3. Male, abdomen, lateral view. × 120
Fig. 4. Male, abdomen, other specimen, lateral view. × 120
Fig. 5. Male, 2nd antenna. × 240

Figs. 6—12. *Corycaeus* (*Onychocorycaeus*) *catus* F. Dahl

Fig. 6. Female, lateral view. × 45
Fig. 7. Female, abdomen, dorsal view. × 60
Fig. 8. Female, 2nd antenna. × 173
Fig. 9. Female, 4th leg. × 173
Fig. 10. Male, dorsal view. × 60
Fig. 11. Male, abdomen, lateral view. × 120
Fig. 12. Male, apical spine of exopodite of 2nd leg. × 200

Figs. 13—20. *Corycaeus* (*Onychocorycaeus*) *pacificus* F. Dahl

Fig. 13. Female, dorsal view. × 30
Fig. 14. Female, abdomen, lateral view. × 45
Fig. 15. Female, 2nd antenna. × 173
Fig. 16. Female, 4th leg. × 173
Fig. 17. Female, apical spine of exopodite of 2nd leg. × 200
Fig. 18. Male, abdomen, lateral view. × 60
Fig. 19. Male, 2nd antenna. × 120

Plate 10.

Figs. 1—5. *Corycaeus* (*Corycella*) *rostratus* Claus

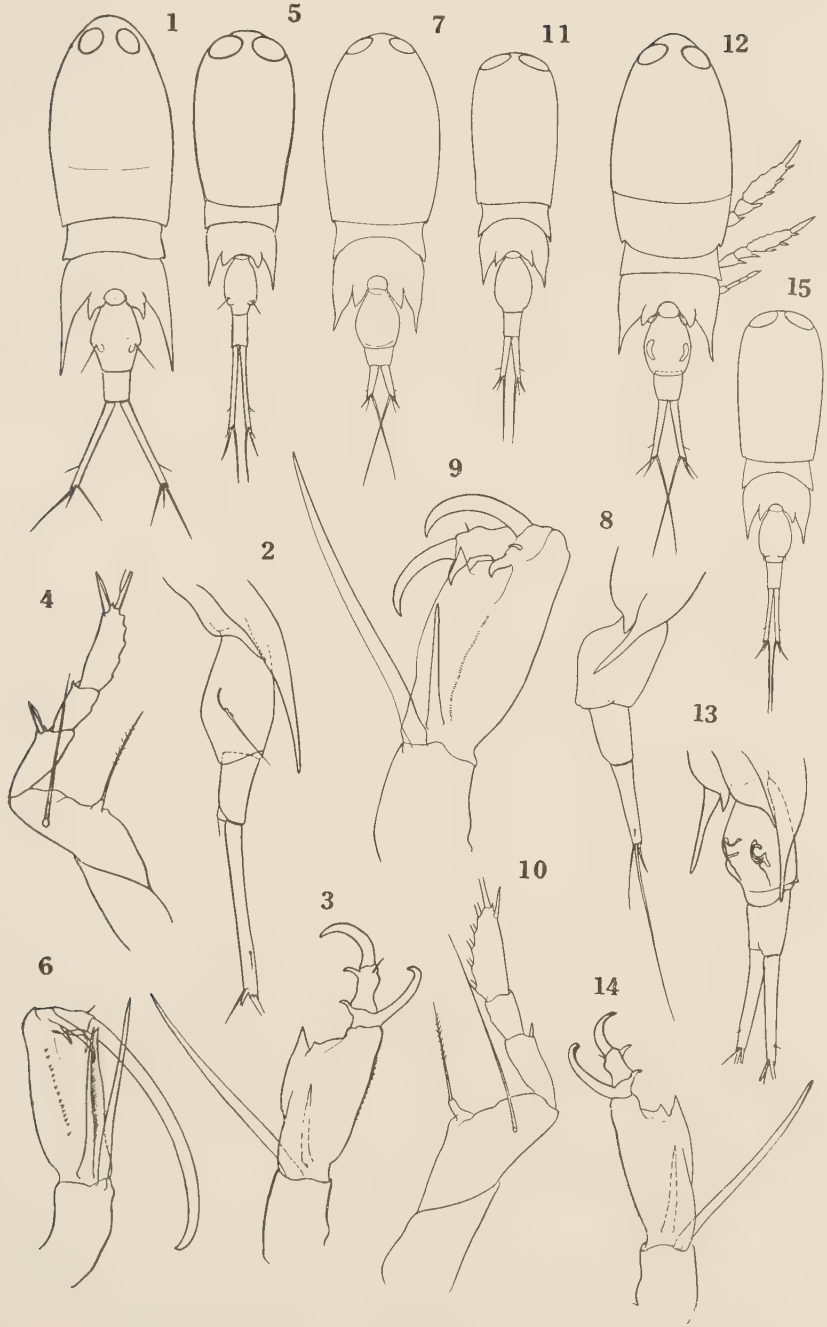
Fig. 1. Male, lateral view. × 83
Fig. 2. Male, cephalothorax, dorsal view. × 120
Fig. 3. Male, abdomen, ventral view. × 120
Fig. 4. Male, 2nd antenna. × 240
Fig. 5. Male, 4th leg. × 353

Figs. 6—11. *Corycaeus* (*Corycella*) *gibbulus* Giesbrecht

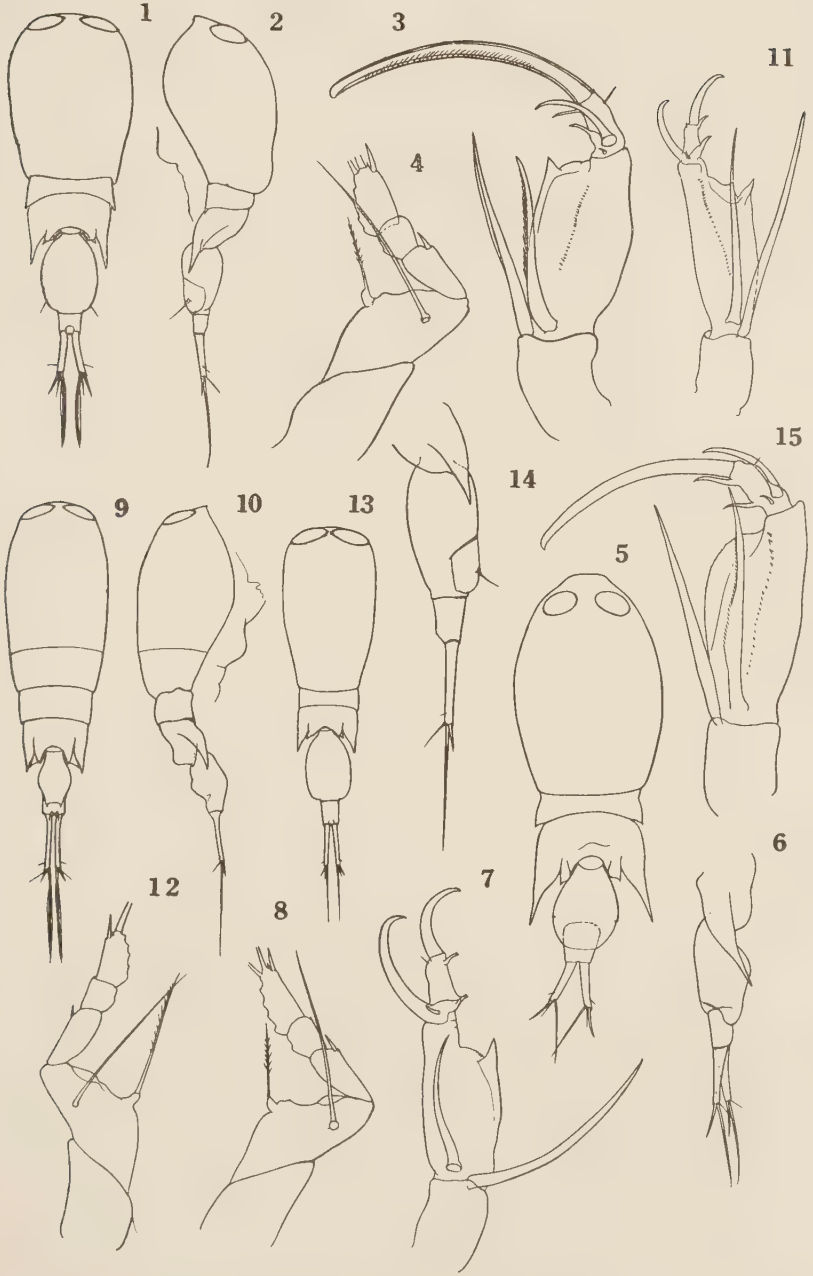
Fig. 6. Female, lateral view. × 45
Fig. 7. Female, abdomen, ventral view. × 60
Fig. 8. Female, 2nd antenna. × 240
Fig. 6. Male, dorsal view. × 60
Fig. 10. Male, abdomen, lateral view. × 120
Fig. 11. Male, 2nd antenna. × 240

Figs. 12—16. *Corycaeus* (*Corycella*) *concinnus* Dana

Fig. 12. Female, dorsal view. × 60
Fig. 13. Female, lateral view. × 60
Fig. 14. Female, 2nd antenna. × 240
Fig. 15. Male, dorsal view. × 60
Fig. 16. Male, abdomen, lateral view. × 120



On Copepoda of the family Corycaeidae



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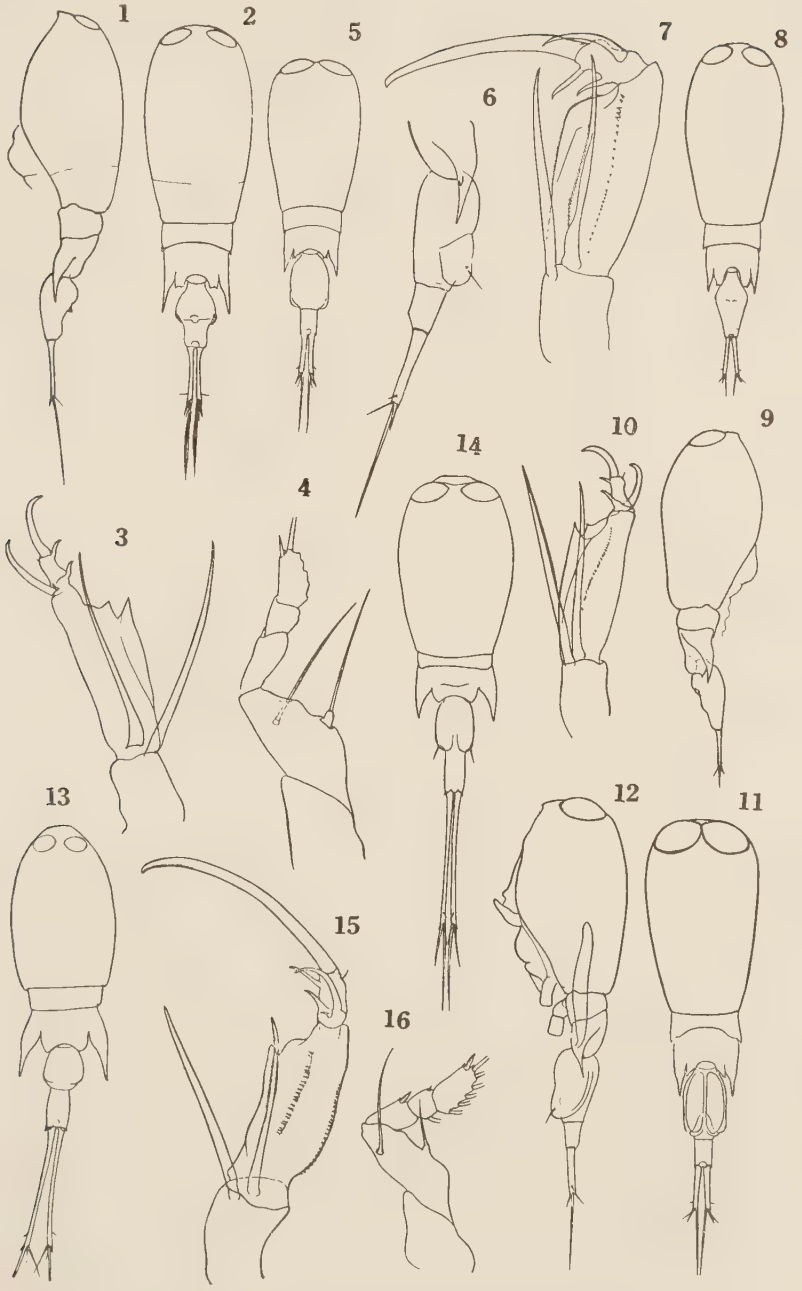

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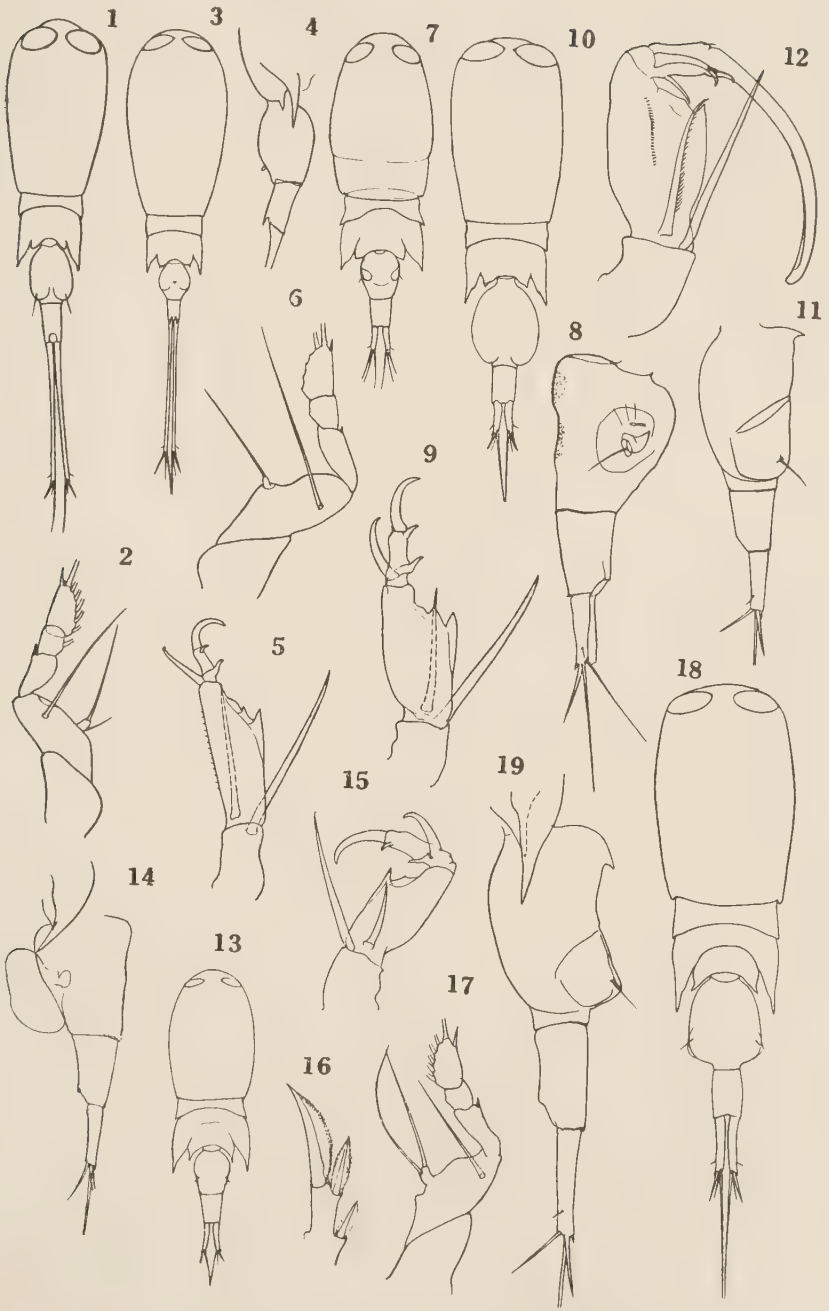

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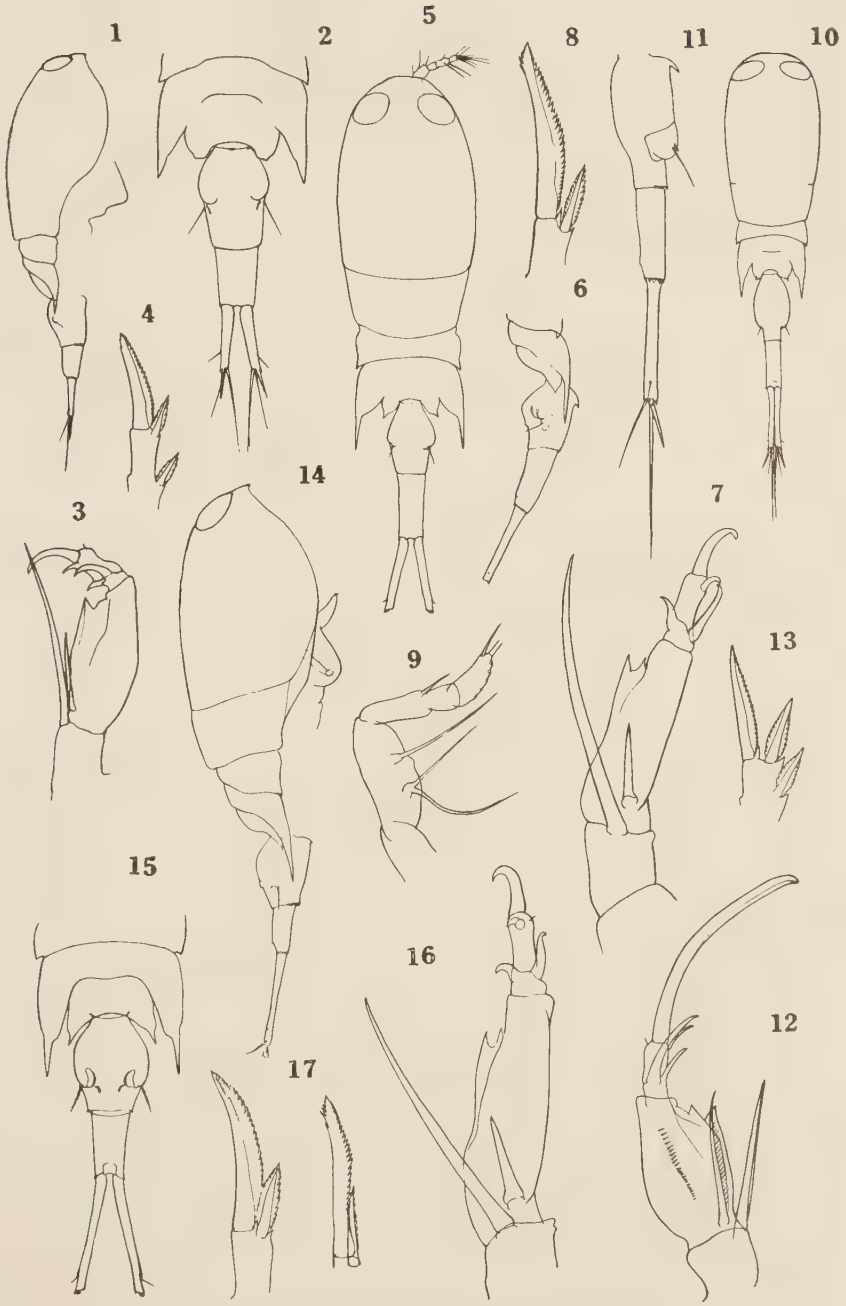

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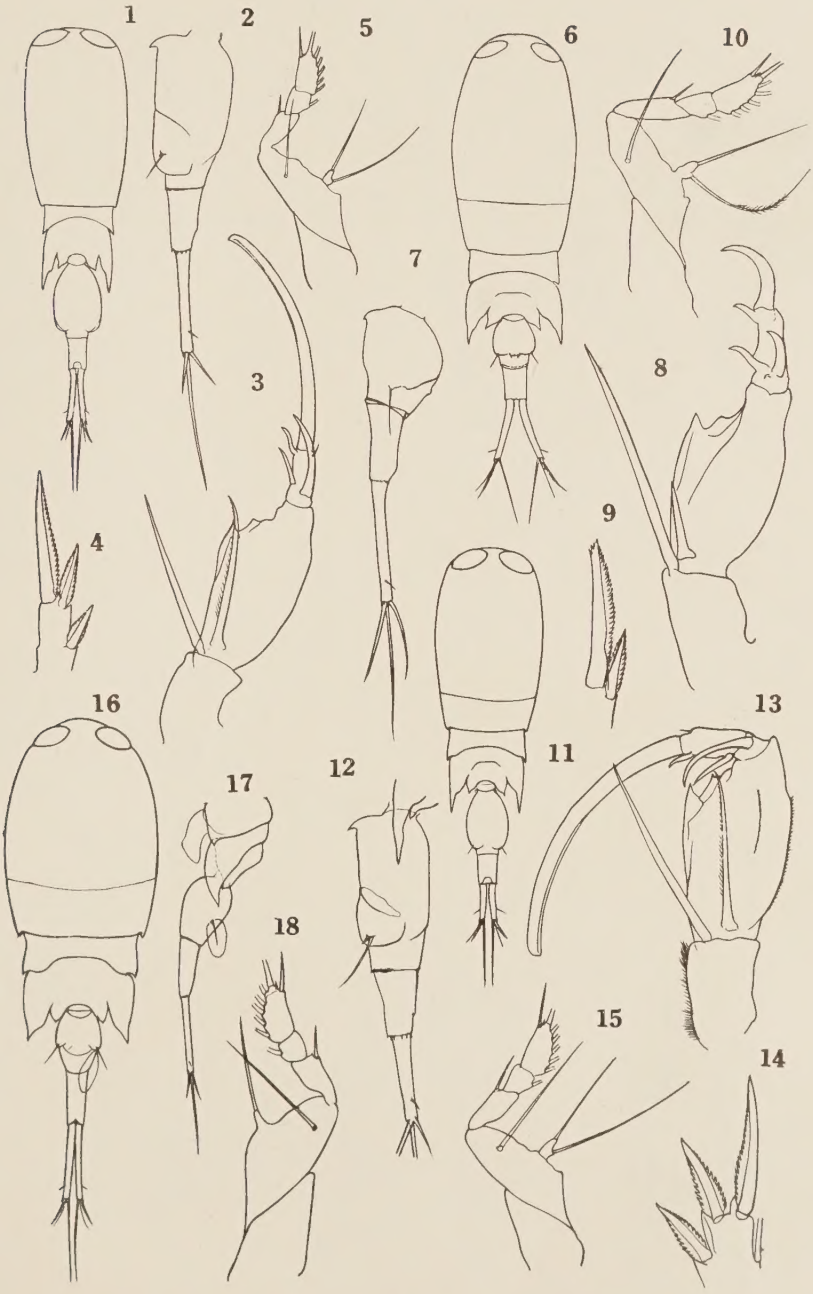

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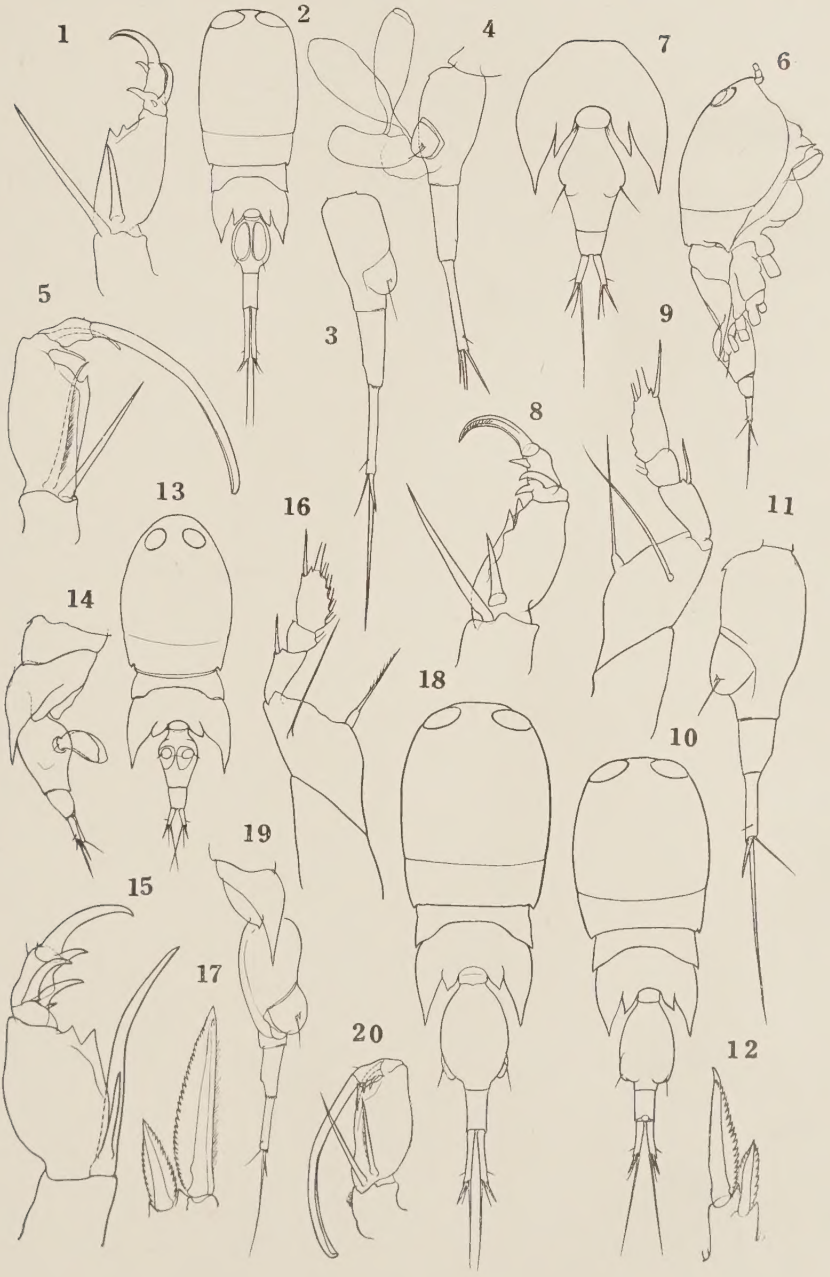

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